SYNTHESIS, CHARACTERIZATION AND BIOLOGICAL ACTIVITY OF 1,3,4- OXADIAZOLE AND 1,3,4- THIADIAZOLE DERIVATIVES

Dissertation Submitted to

The TamilNadu Dr. M.G.R Medical University, Chennai

In partial fulfillment for the requirement of the Degree of

MASTER OF PHARMACY

(Pharmaceutical chemistry)

April - 2012

Department of Pharmaceutical chemistry

KMCH COLLEGE OF PHARMACY

KOVAI ESTATE, KALAPATTI ROAD

COIMBATORE- 641048.

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SMYLIN AJITHA RANI.S

Under the guidance of

Mr. I.PONNILAVARASAN, M. Pharm.

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CERTIFICATE

This is to certify that the dissertation work entitled **"SYNTHESIS, CHARACTERIZATION AND BIOLOGICAL ACTIVITY OF 1,3,4- OXADIAZOLE AND 1,3,4- THIADIAZOLE DERIVATIVES"** submitted by **Mrs.Smylin Ajitha Rani.S.** is a bonafide work carried out by the candidate under the guidance of **Mr. I. Ponnilavarasan M.Pharm,** to the Tamilnadu Dr. M.G.R. Medical University, Chennai, in partial fulfillment for the Degree of **Master of Pharmacy** in Pharmaceutical Chemistry at the Department of Pharmaceutical Chemistry, KMCH College of Pharmacy, Coimbatore, during the academic year 2011-2012.

DATE: **Dr. A. Rajasekaran, M. Pharm., Ph.D.**

PLACE: **Principal**

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Mr. I. Ponnilavarasan, M. Pharm.

DECLARATION

I do here by declare that the dissertation work entitled **"SYNTHESIS, CHARACTERIZATION AND BIOLOGICAL ACTIVITY OF 1,3,4-OXADIAZOLE AND 1,3,4- THIADIAZOLE DERIVATIVES"** submitted to the Tamilnadu Dr. M.G.R. Medical University, Chennai, in partial fulfillment for the Degree of Master of Pharmacy in Pharmaceutical Chemistry, was done by me under the guidance of Prof. I. Ponnilavarasan, M.Pharm, at the Department of Pharmaceutical Chemistry, KMCH College of Pharmacy, Coimbatore, during the academic year 2011-2012.

SMYLIN AJITHA RANI.S

EVALUATION CERTIFICATE

This is to certify that the dissertation work entitled **"SYNTHESIS, CHARACTERIZATION AND EVALUATION OF BIOLOGICAL ACTIVITY OF SUBSTITUTED 1,3,4- OXADIAZOLE and 1,3,4- THIADIAZOLE DERIVATIVES"** submitted by Mrs.Smylin Ajitha Rani.S, University Reg. No: 26107140 to the Tamilnadu Dr. M.G.R. Medical University, Chennai, in partial fulfillment for the Degree of Master of Pharmacy in Pharmaceutical Chemistry is a bonafide work carried out by the candidate at the Department of Pharmaceutical Chemistry, KMCH College of Pharmacy, Coimbatore and was evaluated by us during the academic year 2011-2012.

Examination Center: KMCH College of Pharmacy, Coimbatore.

Date:

Internal Examiner **External Examiner**

Convener of Examinations

DEDICATED TO

MY BELOVED SISTER

Mr.JEYASINGH

Mrs. SHALIN ASHA J. SINGH

&

MR.BASIL VICTOR

OJASWINI ANGEL,JESHWIN

OVIYAS ANGEL.GODWIN

&

MY BELOVED GURU

Mr. I. PONNILAVARASAN

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My dissertation entitled **"SYNTHESIS, CHARACTERIZATION AND BIOLOGICAL ACTIVITY OF 1,3,4-OXADIAZOLE DERIVATIVES AND 1,3,4-THIADIAZOLE DERIVATIVES"** would not have been a feasible one without the grace of ***GOD*** almighty who gave me moral till the completion of my project**.**

I submit my sincere thanks and respectful regard to our beloved Chairman, **Dr. Nalla G. Palanisami** and Managing Trustee, **Dr. Thavamani D. Palanisami** for all the facilities that were provided to me at the institution enabling me to do the work of this magnitude.

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SMYLIN AJITHA RANI.S

ABBREVIATIONS

e.g.	Example
i.e.	That is
%	Percentage
^{1}HNMR	Nuclear Magnetic Resonance
mg	Milligram
ml	Milliliter
µg	Microgram
mm	millimeter
mmol	millimole
w/w	Weight by weight
v/v	Volume by volume
µg/ml	Microgram per litre
hrs	Hours
^{0}C	Degree centigrade
M.P	Melting point
Fig.	Figure
Tab.	Table
UV-VIS	Ultraviolet and visible spectroscopy
min.	Minutes
RBF	Round bottom flask
CCl_4	Carbon tetra chloride
DMF	Dimethyl formamide
DMSO	Dimethyl sulfoxide
IR	Infrared spectoscopy
PPT	Precipitate
M/Z	Mass to charge ratio
TLC	Thin layer chromatography

S.No.	Contents	Pg.no

Abstract:

Various substituted 1,3,4-oxadiazole and 1,3,4-thiadiazole derivatives have been synthesized by cyclising semicarbazone and thiosemicarbazone in presence of ferric chloride and citric acid to form 2-amino-5-substituted 1,3,4-oxadiazole and 1,3,4-thiadiazole respectively. These derivatives undergoes mannich reaction with substituted aniline and formaldehyde to form 2-amino5-substituted1,3,4-oxadiazole and 1,3,4- thiadiazole derivatives. The identity of compounds were confirmed on the basis of their spectral(IR,1H NMR and MASS) data. Further, they have been screened for their antibacterial, antifungal, anticancer and antitubercular activities.

Keywords:

1,3,4-oxadiazoles,1,3,4-thiadiazoles,antibacterial,antifungal,antitubercular,anticancer.

INTRODUCTION

INTRODUCTION

Pharmaceutical chemistry is a science that makes use of the general laws of chemistry to study drugs, i.e. their proportion, chemical nature, composition, influence on an organism and studies of the physical and chemical proportion of drugs, the methods of quality control and the conditions of their usages. In other words ,it is the chemistry of drugs. Drugs mainly exert action depending upon the biochemical pathway.

Pharmaceutical chemistry is a specialized science which depends on the other disciplines such as inorganic, organic, analytical, physical and colloid chemistry and also on medico-biological disciplines such as pharmacology, physiology, biological chemistry etc.

Pharmaceutical chemistry occupies the most important place among the related sciences such as drug technology, toxicology chemistry, pharmacognosy, the economy and the organization of pharmacy.

The very breadth of knowledge required by a medicinal chemist is both a challenge and a reward. Mastering and understanding of such a breadth of subject areas is no straight forward task, but by the same token there is ample intellectual stimulation in understanding the battle against disease at the molecular level and in designing molecular ′soldiers′ to win the battle.

Molecular biology and genetic engineering have produced a deluge of potential new target for drug design and have unraveled the structures and mechanisms of traditional targets while advances in computers and computer aided design have allowed medicinal chemists to take full advantage of this newly earned knowledge.

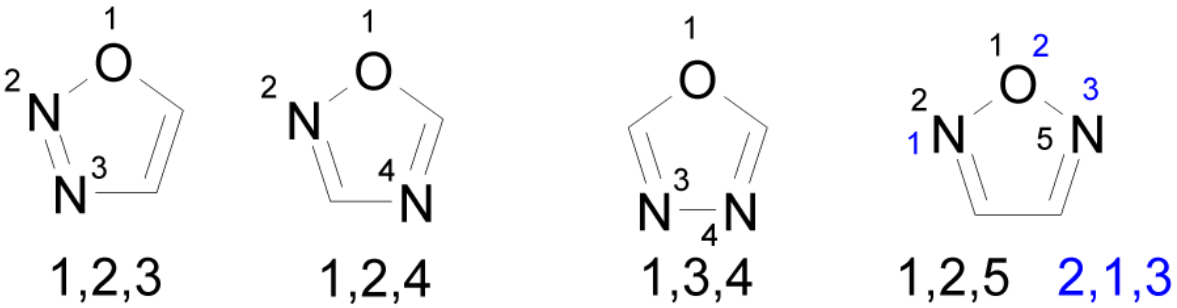
Chemical modification of drug molecules to locate the number of series having optimal effects and will probably continued to be the factor necessary, to make new drugs. To establish the structure of the drug molecules the new invention in the physico-chemical directions such as X-ray crystallography different types of chromatography, spectroscopic studies like NMR, IR, Mass, U.V immensely helpful for medicinal chemist.

The advances in the molecular biology, computer science, instrumentation technology gave an revolutionary turn to concept of chemotherapy leading to development or other area of drug design, QSAR studies etc.

Oxadiazole derivatives are well known to have number of biological and antimicrobial activities. This also having anti-inflammatory, antitubercular, anticancer and anticonvulsant activities.

Thiadiazole and oxadiazoles were reported to possess various pharmacological activity of clinical impotence. Discovering new drugs has never been a simple matter. In view of above considerations we have selected Tailor made approach of drug design in search of new potent bioactive drug molecules.

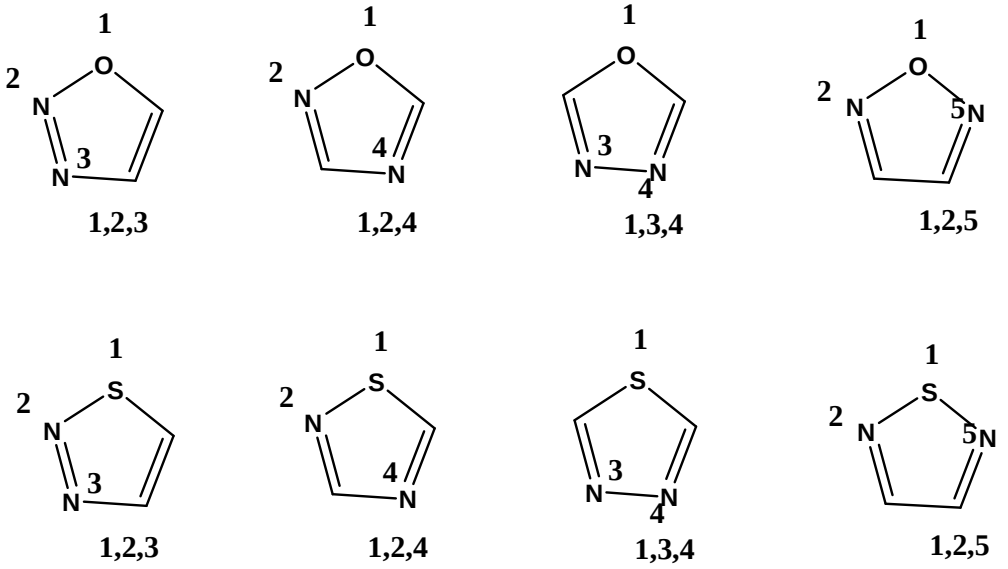
A diversity of useful biological effects is possessed by heterocyclic compounds containing oxadiazole nucleus. Heterocyclic Compounds having a five member ring containing one oxygen and two Nitrogen are called Oxadiazole or in the older literature Furadiazole. **[1]**



In Oxadiazole class 1,3,4- oxadiazole has gradually become prevalent and was used exclusively.

1,3,4-oxadiazoles having many biological activities including, Antioedema and Anti-inflammatory activity**[2,3,4,5],** Analgesic**[3,4]** Antimicrobial**[6,7]** Antitubercular**[8]** Antifungal, Anticonvulsant[9] , Antihypoglycemic, Anticancer[**10,11]**, Antileishmanial, Antimalarial, Antiviral, , and Insecticidal properties.A literature survey also reveals that several 1, 3, 4-oxadiazole derivatives are active against Mycobacterium. The current work describes the synthesis of 1,3,4-oxadiazoles and evaluates the Antibacterial, Antifungal and Antimycobacterial activity against *M. tuberculosis* H37Rv.

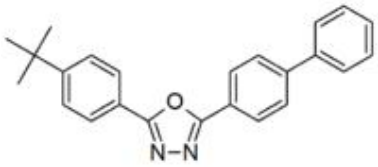
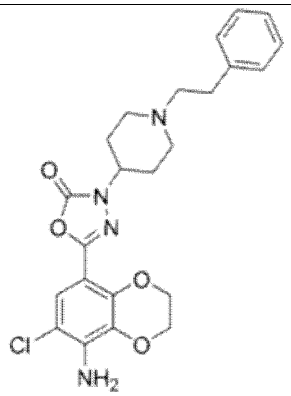
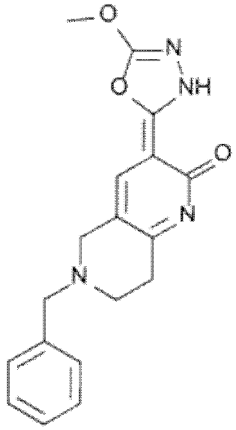
SAR for 1,3,4- Oxadiazole and 1,3,4-thiadiazole



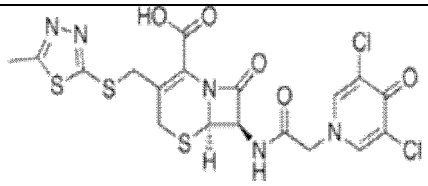
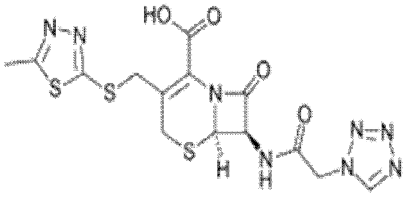
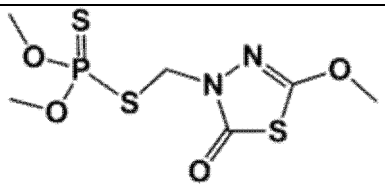
Exhaustive SAR studies have been conducted with the 1, 3, 4-oxadiazole and 1,3,4-thiadiazole derivatives.

- The 2-position and 5-position is an extremely important site of molecular modification, which play a dominant role in determining the pharmacological activities of 1, 3, 4-oxadiazole and 1,3,4- thiadiazole derivative.
- Direct substitution of the 2-position with a 2-methoxy phenol, pyridine and benzoic acid, with pyridine in 5-position enhance the antimicrobial activity of 1, 3, 4-oxadiazole and 1,3,4-thiadiazole derivative.
- Substitution of the 2-position with an methoxybenzene, ethenylbenzene and benzoic acid, with pyridine in 5-position enhance the analgesic activity of 1, 3, 4-oxadiazole and 1,3,4-thiadiazole derivative.
- Direct substitution of the 2-position with an amino benzene and benzoic acid, with pyridine in 5-Position enhance the anti-inflammatory activity of 1, 3, 4-oxadiazole and 1,3,4-thiadiazole derivative.
- Direct substitution of the 2-position with a 2-methoxy phenol, pyridine and benzoic acid, with pyridine in 5-position enhance the Tuberculostatic Activity of 1, 3, 4-oxadiazole and 1,3,4-thiadiazole derivative.

1,3,4-OXADIAZOLE as medicinal agent

Code	Name	Structure	IUPAC	Uses
1	**Butyl PBD**		2-(4-*tert*-Butylphenyl)-5-(4-phenylphenyl)-1,3,4-oxadiazole	In the liquid scintillator neutrino detector.
2	**SL65.0155**		5-(5-amino-6-chloro-2,3-dihydro-1,4-benzodioxin-8-yl)-3-(1-phenethyl-4-piperidyl)-1,3,4-oxadiazol-2-one hydrochloride	is a selective 5-HT_4 receptor partial agonist. It potently enhance cognition,learning,memory and also possesses antidepressant effects.
3	**SX-3228**		(3*E*)-6-benzyl-3-(5-methoxy-1,3,4-oxadiazol-2(3*H*)-ylidene)-5,6,7,8-tetrahydro-1,6-naphthyridin-2(3*H*)-one	Is a sedative and hypnotic and with only limited anxiolytic effects which appear only at doses that also produce significant sedation.

1,3,4-THIADIAZOLE as medicinal agent

Code	Name	Structure	IUPAC	Uses
1	**Cefazedone**		(6*R*,7*R*)-7-{[2-(3,5-dichloro-4-oxopyridin-1-yl) acetyl]amino}-3-[(5-methyl-1,3,4-thiadiazol-2-yl) sulfanylmethyl]-8-oxo-5-thia-1-azabicyclo[4.2.0] oct-2-ene-2-carboxylic acid	It is used in the treatment of respiratory and urinary tract infections
2	**Cefazolin**		(6*R*,7*R*)-3-{[(5-methyl-1,3,4-thiadiazol-2-yl)thio]methyl}-8-oxo-7-[(1*H*-tetrazol-1-ylacetyl)amino]-5-thia-1-azabicyclo[4.2.0]oct-2-ene-2-carboxylic acid	Cefazolin is mainly used to treat bacterial infections of the skin. It can also be used to treat moderately severe bacterial infections involving the lung, bone, joint, stomach, blood, heart valve, and urinary tract.
3	**Methidathion**		3-(dimethoxyphosphinothioylsulfanylmethyl)-5-methoxy-1,3,4-thiadiazol-2-one	is an organophosphate insecticide.

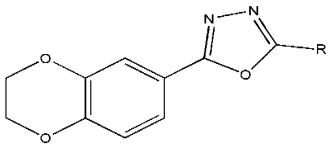
LITERATURE REVIEW

LITERATURE REVIEW

In the family of Heterocyclic compounds, containing Nitrogen, Oxygen atom and Sulphuratom are considered as an important class of compounds in Medicinal Chemistry because of their interesting diversified Biological application. During the past years considerable evidences have also accumulated to demonstrate the efficacy of 1, 3, 4-oxadiazoles and 1,3,4-thiadiazoles including Antibacterial, Anti-inflammatory, Anti malarial, Antitubercular, Anti hypoglycemic, Anticancer, Antileishmanial, Antialzheimer, Antiviral, Anticonvulsant and Insecticidal properties. Literature survey revealed that slight modification in the structure can result in qualitative as well as quantitative changes in the activity, which prompted us to undertake the synthesis of various new 2-amino-5-substituted-1, 3, 4-Oxadiazole derivatives with the aim of having improved activity and lesser toxicity. The review of literature reveal also prompted us to synthesize substituted Oxadiazole and thiadiazole compounds and those will be screened for Antimicrobial activity to get potent bioactive molecule. It is well known that introduction of atom like chlorine, bromine, nitro etc in organic molecule causes dramatic change in its biological profile.The synthesized compound will be screened for anticancer activity and antitubercular activity.

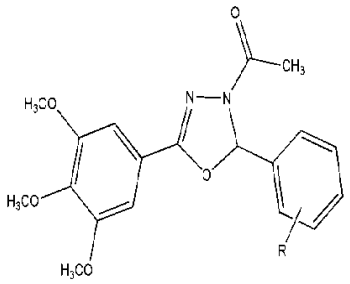
Oxadiazole as Anticancer agents

Xiao-Min Zhang etal[13] synthesized a series of novel 1,3,4-oxadiazole derivatives containing 1,4-benzodioxan moiety (6a–6s) as potential telomerase inhibitors were synthesized. compound 6k possessed the most potent telomerase activity .



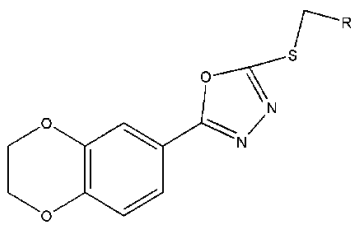
6K=2-Me-C6H4

Linhong Jin,etal[14] synthesized a series of 3-acetyl-2-substituted phenyl-5-(3,4,5-trimethoxyphenyl)-2,3-dihydro-1,3,4-oxadiazole derivatives . The IC50 values of high active compounds 2a, 2b, 2c, 2f, 3l, and 3m against PC3 cells and are moderately active against Bcap37 and BGC823 cells.



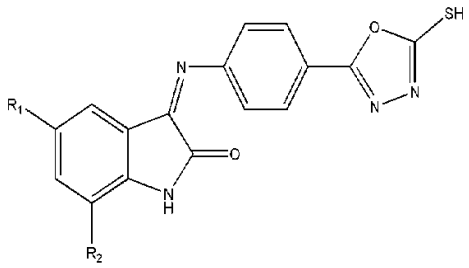
R=a:2-F, b:3-F,c:4-F,d:2-CF_3, f 4CF_3, l:3,5-2Cl, m:2,4-3OCH_3

Hai-Liang Zhu etal[15] synthesized a series of a series of new 2-chloropyridine derivatives possessing 1,3,4-oxadiazole moiety were synthesized. Antiproliferative assay results indicated that compounds 6o and 6u exhibited the most potent activity against gastric cancer cell which was more potent than the positive control.



Where6k=2-Me-C6H4

Rajyalakshmi Gudipati etal[16] synthesized A series of 5- or 7-substituted 3-{4-(5-mercapto-1,3,4-oxadiazol-2-yl)phenylimino}-indolin 2-one derivatives were synthesized by treating 5-(4-aminophenyl)-1,3,4-oxadiazole-2-thiol.



Xiaoling Chen etal[17] synthesized 1,3,4- Oxadiazole derivatives were synthesized and evaluated for their ability to inhibit tubulin polymerization and to cause mitotic arrest in tumor cell.

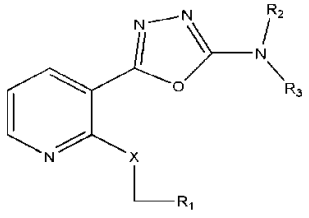
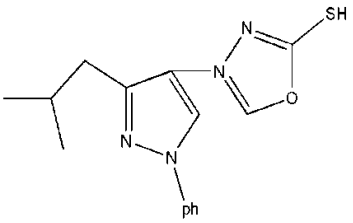


Figure R^1=4-Fluorophenyl,R^2=2,3-dihydro benzo-1,4-dioxin-6yl,R^3=H shows activity in tumour cell lines.

M.A. Abu-Zaie etal [18] synthesized of novel 1,3,4-oxadiazole condense with O-aminothiols to give the bases 8, 19 and 20 in good yields, respectively. Pharmacological evaluation of compounds 8, 14, 16 and 22 in vitro against 2-cell lines MCF-7 (breast) and HEPG2 (liver) revealed them to possess high anti-tumor activities with IC50 values ranging from 2.67e20.25 (mg/mL) for breast cell line (MCF-7) and 4.62e43.6 (mg/mL) for liver cell line (HEPG2).



Xiaohu Ouyang et.al[19] Synthesized a series of 1, 3, 4 oxadiazoles and are evaluated for their ability to inhibit tubulin polymerization and to arrest mitotic division of tumor cells. Among these synthesized compounds, some compound showed potent activity.

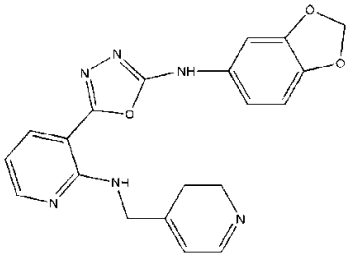


Fig: 3-(5-(benzo[d][1, 3]dioxol-5-ylamino)-1,3,4-oxadiazol-2-yl)-N-(pyridin-4-ylmethyl)pyridin-2-amine

B Shivarama holla et.al[20] Synthesized 2chloro 1,4 bis-(5-sustituted 1,3,4 oxadiazole-2-ylmethylenoxy) phenylene derivative and these compound have been extensively screened against a panel of sixty cancer cell lines derived from leukemia, prostrate, colon , lung, CNS, ovarian, melanoma and breast cancer respectively. Compound 3d and 3f shows potent Anticancer activity against most of the cell lines with GI50 values <s100 μM concentrations.

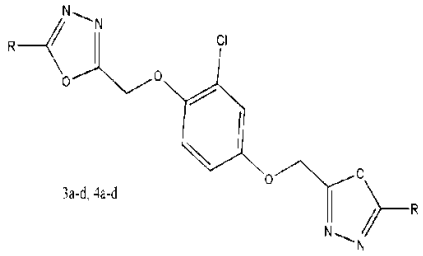


Fig: 2chloro 1, 4 bis-(5-sustituted 1, 3, 4 oxadiazole-2-ylmethylenoxy) phenylene derivatives

3a	$4ClC_6H_4$
3b	$4NO_2C_6H_4$
3c	$4ClC_6H_4OCH_2$
3d	$2,4Cl_2C_6H_3OCH_2$
4a	$4ClC_6H_4$-furanyl
4b	$4NO_2\ C_6H_4$-furanyl
4c	$2,4Cl_2\ C_6H_3$-furanyl
4d	$4\text{-}NO_2\text{-}2\text{-}CH_2\ C_6H_3$-furanyl

Maryam Zahid etal[21] Synthesized a series of Adamantanyl-1, 3-thiazole and 1,3,4-oxadiazole derivatives (**6a-l**), bearing various aryl groups has been synthesized from adamantan-1-nitrile in four steps. All the compounds were evaluated, in vitro, for Anti proliferative activity against a large panel of human tumor-derived cell lines. Compounds **6e** exhibited activity against human spleenic lymphoblastoid (WIL-2NS) and human acute B-lymphoblastic leukemia (CCRF-SB) cell lines with CC_{50} = 68 and 42μM, respectively. Compound **6l** showed activity against CCRF-SB cell lines with CC_{50} = 51 μM. All the other compounds were found inactive.

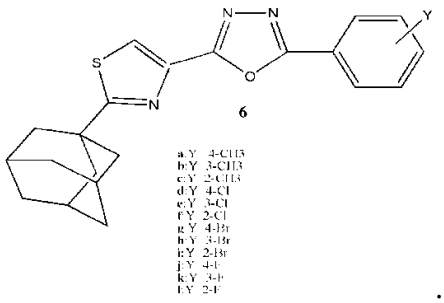


FIG: 2-(2-adamantyl-1, 3-thiazol-4-yl)-5-aryl-1, 3, 4-oxadiazoles.

Aboraia AS et al[22] Synthesized a series of 5-(2-hydroxyphenyl)-3-substituted-2,3-dihydro-1,3,4-oxadiazole-2-thione derivatives were evaluated for their in-vitro Anticancer activity, where seven out of twenty two synthesized compounds displayed high Anticancer activity, in the primary assay. These seven oxadiazole compounds were selected for a full Anticancer screening against a 60-cell panel assay where they showed non-selective broad spectrum and

promising activity against all Cancer cell lines. As a result of 60-cell panel assay two oxadiazole compounds were identified as promising lead compounds.

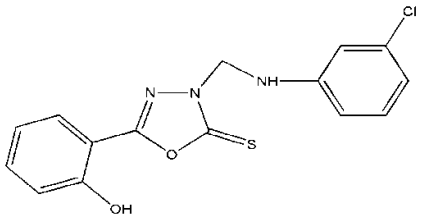
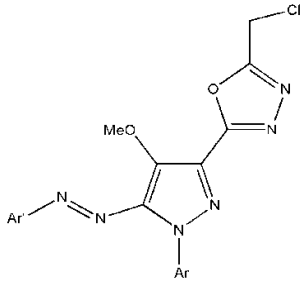


Fig: 3-((3-chlorophenylamino)methyl)-5-(2-hydroxyphenyl)-1,3,4-oxadiazole-2(3H)-thione

Oxadiazole as AntiDiabetic

Hanna et al [23] **S**ynthesized a series of novel 1, 3, 4 oxadiazole derivatives and evaluated for hypoglycemic activity and the synthesized compounds have good hypoglycemic activity.



Hussain et al [24] **S**ynthesized a series of 2-arylamino-5-(2 naphthyloxymethyl)-1, 3, 4-oxadiazole derivatives has exhibited considerable Oral Hypoglycemic activity.

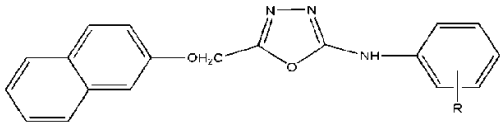


Fig: 2-arylamino-5-(2-naphthyloxymethyl)-1, 3, 4-oxadiazole derivative

Husain MK and Jamali MR, [25] **S**ynthesized a series of 1, 3, 4-oxadiazole analogues *i.e.*, 2-arylamino-3-*p*-(3-aryl-4-oxaquinozolin-2-yl (methylamino) phenyl-1,3,4-oxadiazoles has been found to possess Oral Hypoglycemic activity.

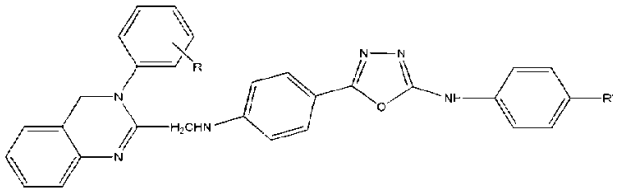
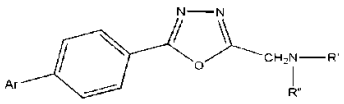


Fig: 2-arylamino-3-*p*-(3-aryl-4-oxaquinozolin-2-yl (methylamino) phenyl-1, 3, 4-oxadiazoles

A series of 2, 5-disubstituted 1, 3, 4-oxadiazoles derivatives with different Substituent's were synthesized for their hypoglycemic activity



Oxadiazole as Anticonvulsant

Omar Amme and Aboulwafa om,[26] Synthesized a novel series of 2-substituted amino-5-aryl1, 3, 4-oxadiazole derivatives were synthesized and screened for their anticonvulsant properties.

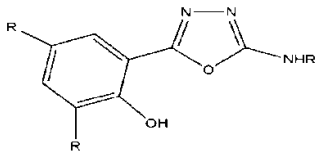


Fig: 2-substituted amino-5-aryl-1, 3, 4-oxadiazol

Zarghi et al[27] Synthesized a series of 2-substituted-5-(2-benzyloxyphenyl)-1, 3, 4-oxadiazole derivatives were synthesized and evaluated as Anticonvulsant agent.

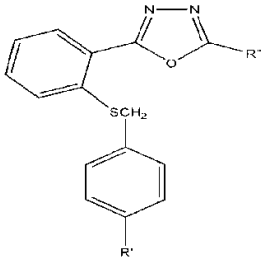


Fig: 2-substituted-5-(2-benzyloxyphenyl)-1, 3, 4-oxadiazole derivatives

Choudhary etal[28] synthesized some Mannich bases like 5-(3,4-methylenedioxyphenyl)-3-arylaminomethyl-1,3,4-oxadiazole-2-thiones, were synthesized for their Anticonvulsant properties.

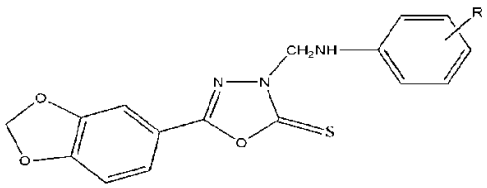
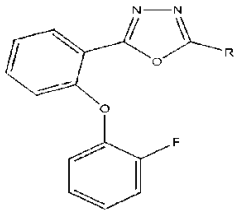
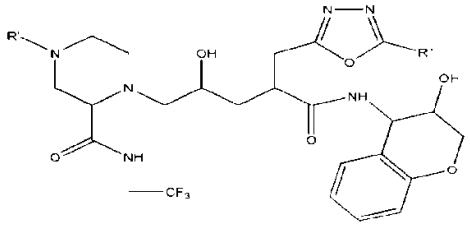


Fig: 5-(3, 4-methylenedioxyphenyl)-3-arylaminomethyl-1, 3, 4-oxadiazole-2-thiones

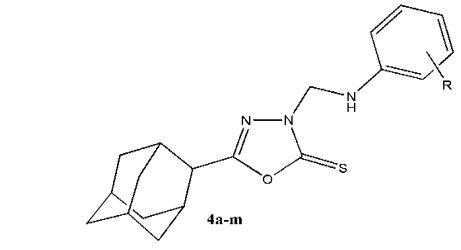
Almasirad A etal[29] synthesized some of the compounds i.e., 2-substituted-5-[2-(2-fluorophenoxy) phenyl]-1, 3, 4-oxadiazoles and -1, 2, 4-triazoles showed considerable Anticonvulsant activity in PTZ and MES models.



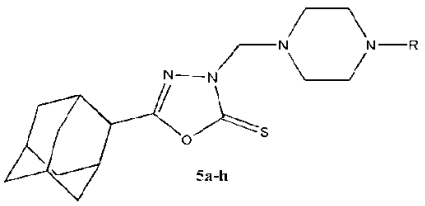
Kim RM etal[30] Synthesized a series of HIV-1 protease inhibitors (PI's) bearing 1,3,4-oxadiazoles at the P1' position were synthesized by a novel method involving the Diastereoselective installtion of a carboxylic acid and conversion to the P1' heterocycle exhibited excellent activities.



Ali A. El-Emam[31] Synthesized a series of 5-(1-adamantyl)-3-arylaminomethyl- 1,3,4-oxadiazoline-2-thiones 4a–m or 5-(1-adamantyl)-3-(4-substituted-1-piperazinylmethyl)-1,3,4-oxadiazoline-2-thiones 5a–h, The inhibitory activity of compounds 2, 4a–m, and 5a–h against Human Immunodeficiency virus type 1 (HIV-1) was determined using the XTT assay in MT-4 cells.



4a-m



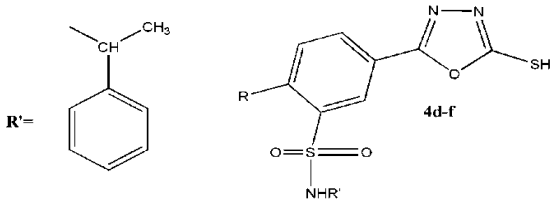
5a-h

4a=H
4b=2-F
4c=4-F
4d=2-Cl
4e=4-Cl
4f=2-Br
4g=4-Br
4h=3-NO_2
4i=4-NO_2
4j=4-CH_3O
4k=2-CN
4l=2-CF_3
4m=2,5-F2

5a=CH_3
5b=C_2H_5
5c=$COOC_2H_5$
5d=C_6H_5
5e=4-FC_6H_4
5f=2-$CH_3OC_6H_4$
5g=$C_6H_5CH_2$
5h=2-$CF_3C_6H_4CH_2$

Fig: 5-(1-adamantyl)-3-arylaminomethyl-1,3,4-oxadiazoline-2-thiones 4a–m or 5-(1-adamantyl)-3-(4-substituted-1-piperazinylmethyl)-1,3,4-oxadiazoline-2-thiones 5a–h,

R. Iqbal etal[32] Synthesized Twelve novel primary and secondary Benzene sulfonamides bearing 2,5-disubstituted-1,3,4-oxadiazole moiety. Some of the synthesized compounds (**4d, 4e, 4f**) have been screened in vitro for their anti-HIV activity; the results were in accordance with SAR.



R'=

4d-f

4d:R=H, **4e:**R=OCH3, **4f:**R=Cl

Fig: 3-(5-mercapto-1, 3, 4-oxadiazol-2-yl) benzene sulfonamide derivatives.

Oxadiazole as Analgesic and Anti-inflammatory

Jayashankar etal.[33] Synthesized a series of novel ether- linked bis(heterocycle)s. all the synthesized compounds were screened for Anti-inflammatory and analgesic activities,7d and 7g showed excellent activity than Ibuprofen and Aspirin.

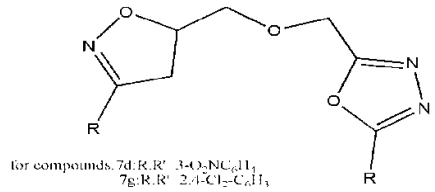


Fig: 2-(((4, 5-dihydroisoxazol-5-yl)methoxy)methyl)-1,3,4-oxadiazole

Shashikant V. Bhandari etal[34] Synthesized a series of S- substituted phenacryl 1,3,4 oxadiazole and Schiff bases derived from 2-[(2,6-dicloroanilino)phenyl acetic acid], (Diclofenac acid) were found to have significant Anti-inflammatory activity with significant analgesic activity acetic acid induce Writhing models with no Ulcerogenic activity , those 8 active compounds found to have most prominent and consistent Anti-inflammatory activity.

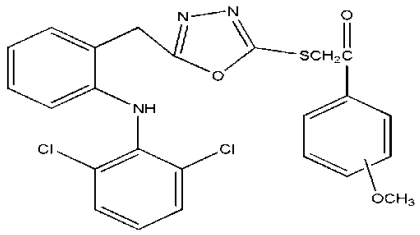


Fig: S-substituted phenacryl 1, 3, 4 oxadiazole and schiff bases derived from 2-[(2, 6-dicloroanilino) phenyl acetic acid]

Mohd Amir etal[35] Synthesized a series of new 1, 3, 4 oxadizole derivatives and 1, 2, 4 triazine-5-one derivatives. All compounds were screened for their Anti-inflammatory activity by using Carrageenin- induced rat paw edema method. Compounds synthesized shown maximum Anti-inflammatory activity.

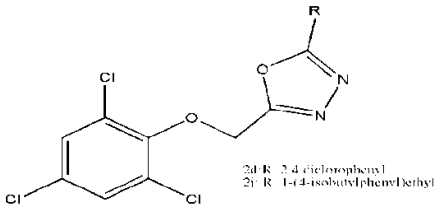


Fig: 2-((2, 4, 6-trichlorophenoxy) methyl)-1,3,4-oxadiazole derivatives.

Harish Rajak, Kramer etal[36] synthesized a series of 1, 3, 4-oxadiazole derivatives have found to exert their anti-inflammatory effect via Cyclooxygenase and 5-Lipoxygenase inhibitory activity.

DTBP = 3,5-di-tert-butyl-4-hydroxyphenyl

Nigam etal[37] Synthesized a series of 2-thio-3-(substituted-amino methyl)-5-(3, 5-dinitrophenyl)-1, 3, 4-oxadiazoles has been found to possess considerable Anti-inflammatory property.

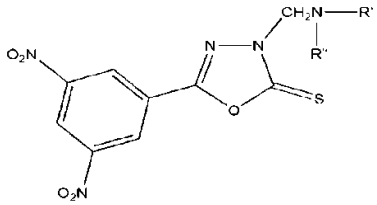


Fig: 2-thio-3-(substituted-aminomethyl)-5-(3,5-dinitrophenyl)-1,3,4-oxadiazoles

Burbulien N etal.[38] Synthesized a series of 5-[(2-disubstitutedamino-6-methyl-pyrimidin-4-yl)-sulfanylmethyl]-3H-1,3,4-oxadiazole-2-thiones among these he found that some of these derivative were much more potent than Ibuprofen.

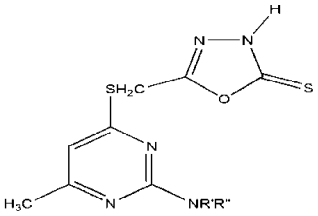
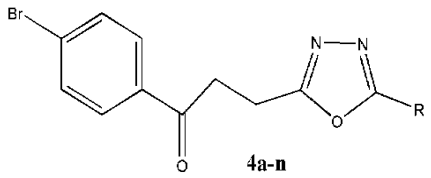


Fig: 5-[(2-disubstitutedamino-6-methyl-pyrimidin-4-yl)-sulfanylmethyl]-3H-1, 3, 4-oxadiazole-2-thiones

Asif HusainMohammed Ajmal etal[39] Synthesized a novel series of 2-[3-(4-bromophenyl)propan-3-one]-5--(substituted phenyl)-1,3,4-oxadiazoles (**4a-n**) have been synthesized from 3-(4 bromobenzoyl)propionic acid with the aim to get better Anti-inflammatory and Analgesic agents with minimum or without side effects (ulcerogenicity).

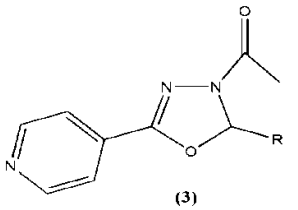


4a-n

R: C_6H_5 -, 2-ClC_6H_4 -, 4-ClC_6H_4 -, 2-HOC_6H_4-, 4-$NO_2C_6H_4$-, 4-FC_6H_4-,4-$CH_3C_6H_4$ -, 4-$OCH_3C_6H_4$ -, 3,4-$(OCH_3)2C_6H_3$-, $C_6H_5CH_2$-,$C_6H_5OCH_2$ -, 2-$C_6H_5COC_6H_4$-, 1-$C_{10}H_7OCH_2$-, 2-$C_{10}H_7OCH_2$-

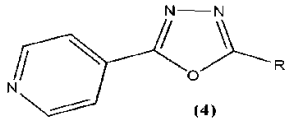
Fig: 2-[3-(4-bromophenyl) propan-3-one]-5--(substituted phenyl)-1, 3, 4-oxadiazoles

Dhansay Dewangan1, Alok Pandey etal[40] Synthesized a series of novel 1,3,4 oxadiazole such as 3 (3a-3e) and 4 (4f-4j) and screened for anti-inflammatory and analgesic activity compound 4g and 4j was found to possess better activity then others.



(3)

3a: R=2-OH C_6H_4
3b: R=4-OCH_3-C_6H_4
3c: R=3-OCH_3-4-OH-C_6H_3
3d: R=2-C_4H_3O
3e: R=H_5C_6

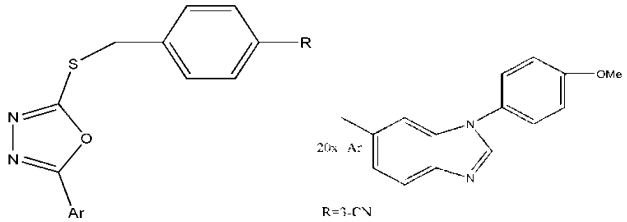


(4)

4f:R=C_8H_8
4g:R=C_5H_4N
4h:R=2-NH_2-C_6H_4
4i:R=4-NH_2-C_6H_4
4j:R=2-COOH-C_6H_4

Oxadiazole as Antialzheimer

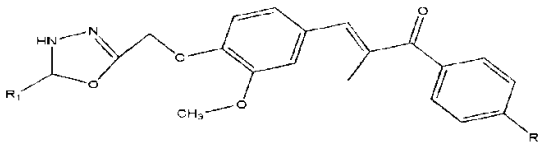
Morihisa Saitoh etal[41] have been reported synthesis and structure–activity relationships of 1,3,4-oxadiazole derivatives as novel inhibitors of glycogen synthase kinase-3b is implicated in abnormal hyperphosphorylation of tau protein and its inhibitors are expected to be a promising therapeutic agents for the treatment of Alzheimer's disease. The synthesized compound 20x showed highly selective and potent GSK-3b inhibitory activity in vitro .



Where

Oxadiazole as Antimicrobial

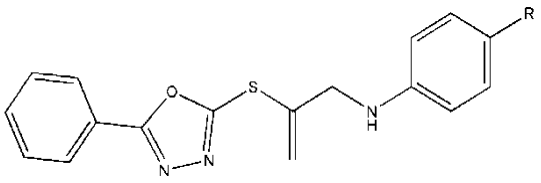
Mohammed Afroz Bakht etal[42] have been reported the molecular properties prediction, synthesis and antimicrobial activity of some newer 1,3,4- oxadiazole derivatives were synthesized and characterized by IR, NMR and mass spectral analysis followed by antibacterial and antifungal screening. It was observed that compounds showed moderate to good antibacterial activity, but their antifungal activity was somewhat moderate. Compounds AB13 and AB20 showed pronounced activity against all bacterial and fungal strains.



Compound No.	R	R'
AB1	H	C_6H_5
AB2	H	p-Cl−C_6H_4
AB5	H	p-CH_3−C_6H_4
AB6	H	o-OH−C_6H_4
AB7	H	C_6H_5−OCH_2
AB8	OCH3	C_6H_5
AB9	OCH3	p-Cl−C_6H_4
AB12	OCH3	p-CH_3−C_6H_4
AB13	OCH3	o-OH−C_6H_4
AB14	OCH3	C_6H_5−OCH_2
AB15	OH	C_6H_5
AB16	OH	p-Cl−C_6H_4
AB18	OH	p-NO_2−C6H4
AB19	OH	p-CH_3−C_6H_4
AB20	OH	o-OH−C_6H_4

Jignesh P. Raval etal[43] synthesized series of 2(4pyridyl)5[(aryl/heteroarylamino)-1-oxoethyl]thio-1,3,4-oxadiazole the synthesized compounds, compounds 2(pyridyl)-5((2-nitrophenylamino)-1-oxoethyl)thio-1,3,4-oxadiazole (5e), 2(4-pyridyl)-5((4-

nitrophenylamino)-1-oxoethyl)thio-1,3,4-oxadiazole (5g) and 2(4-pyridyl)-5((2-pyrrolylamino)-1-oxoethyl)thio-1,3,4-oxadiazole (5k) produced highest efficacy and exhibited >90% inhibition at a concentration of 0.0077, 0.0052 and 0.0089 lM, respectively. All the new compounds were pharmacologically evaluated for their in vitro Antimicrobial activity.



Rakesh Chawla, Anshu Arora etal [44] Synthesized 3-acetyl-5-(3-chloro-1 benzo [b]thiophen-2-yl)-2-substituted phenyl-2,3-dihydro-1,3,4-oxadiazoles and 2-(3-chloro-1-benzo[b] thiophen-2-yl)-5-substituted phenyl-1,3,4-oxadiazole and was evaluated for Antimicrobial activity. Compound 4c and 4e were found to be most potent, even better than standard drug i.e. Ciprofloxacin.

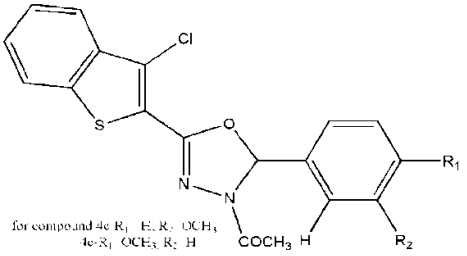
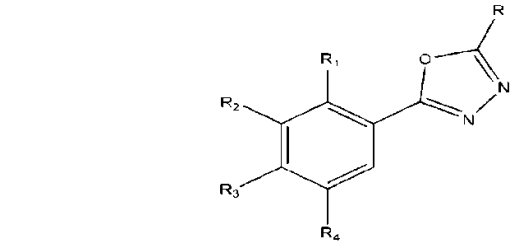


Fig: acetyl-5-(3-chloro-1 benzo [b]thiophen-2-yl)-2-substituted phenyl-2,3-dihydro-1,3,4-oxadiazoles and 2-(3-chloro-1-benzo[b] thiophen-2-yl)-5-substituted phenyl-1,3,4-oxadiazole derivative

Chandrakant etal [45] Synthesized a series of new 1,3,4 oxadiazole with 2-fluoro-4 methoxy moiety and are tested for Antimicrobial activity, Compound 4a and 4b from all synthesized compounds showed significant Antibacterial activity against Escherichia coli and Pseudomonas aeruginosa, 4i showed good Antifungal activity against C. albicans.



for compound 4a:R=2-fluoro-4-methoxyphenyl, R_1=CH_3, R_2=Br, R_3=H, R_4=H
4b: R=2,3,4-trifluorophenyl, R_1=F, R_2=H, R_3=OCH_3, R_4=H
4c: R=2-fluoro-4-methoxyphenyl, R_1=Br, R_2=H, R_3=H, R_4=Cl

Mohamed Ashraf Ali and Mohammad Shaharyar etal [46] Synthesized a series of Oxadiazole Mannich bases by reaction between Oxadiazole derivatives, Dapsone, appropriate Aldehydes and was evaluated against Mycobacterium Tuberculosis. Compound 3-{2-furyl[4-(4-{2-furyl [5-(2-naphthyloxymethyl)-2-thioxo-2,3-dihydro-1,3,4-oxadiazol-3 yl] methylamino} phenylsulfonyl) anilino]methyl}-5-(2-naphthyloxymethyl)-2,3-dihydro-1,3,4-oxadiazole-2-thione from all the synthesized compounds have shown best activity against M. Tuberculosis and Isoniazide resistant M. Tuberculosis.

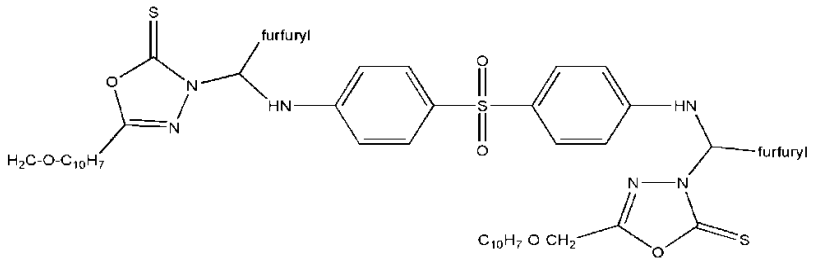


Fig : 3-{2-furyl[4-(4-{2-furyl[5-(2-naphthyloxymethyl)-2thioxo2,3dihydro1,3,4oxadiazole3yl]methylamino} phenylsulfonyl)anilino]methyl}-5-(2-naphthyloxymethyl)-2,3 dihydro-1,3,4-oxadiazole-2-thione

Manish Kumar Mishra etal[47] Synthesized 6 – Methyl – 4 – aryl – 5 - (5- phenyl -1, 3, 4 – oxadiazol -2- yl) -1, 2, 3, 4-tetrahydropyrimidine-2(1H)-one. Among the derivatives 3e has significant effect against Streptococcus pneumonia (+ve) and 3b has significant activity effect Escherichia coli(-ve)

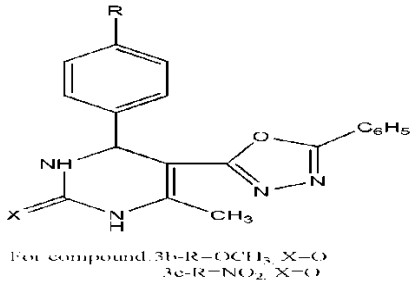


Fig: New 6 - Methyl - 4 - aryl - 5 - (5- phenyl -1, 3, 4 -Oxadiazol -2- yl) -1, 2, 3 , 4-tetrahydropyrimidine-2(1H)-one

M. Shahar Yar etal[48] Synthesized a series of novel 2,5-disubstituted 1,3,4-oxadiazolederivatives and was tested for invitro Anti-Microbial activity. 2-(2 naphthyloxymethyl)-5-phenoxymethyl-1, 3, 4-oxadiazole exhibited > 90% inhibition among all the synthesized compounds.

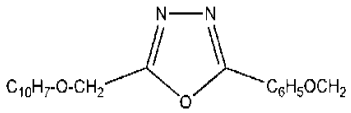


Fig: 2, 5-disubstituted 1, 3, 4-oxadiazole derivatives

Mojahidul Islam etal[49] Synthesized a series of 5-{3'-oxo-6'-(substituted aryl)-2', 3', 4', 5'-tetrahydropyridazin-2i-ylmethyl}-2-substituted 1, 3, 4-oxadiazole and then final compounds were tested for their Anti- bacterial activity using cup plate method. Out of all the synthesized compounds 2e and 4 found to be most potent derivative as compared to the standard drug.

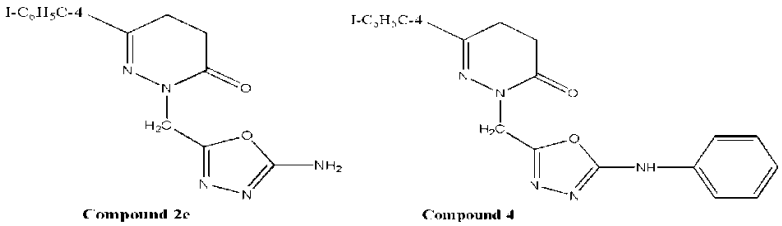
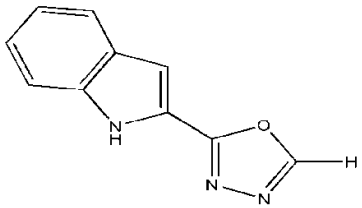


Fig: 5-{3'-oxo-6'-(substituted aryl)-2', 3', 4', 5'- tetrahydropyridazin-2i-ylmethyl}-2-substituted 1, 3, 4oxadiazole

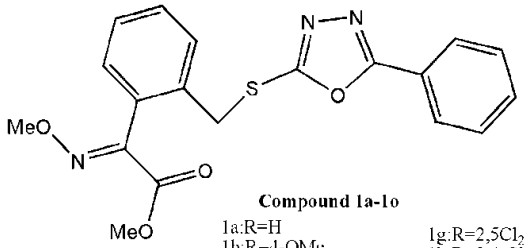
Nitin Bhardwaj etal[50]. Synthesized 1,3,4-oxadiazole from different compounds and were tested for Anti- Microbial activity on different strains. A total of four compounds were

synthesized out of those only three found to be effective against bacterial strains and none of the strains were found to be effective against fungal strain.



2-(1,3,4-oxadiazol-2-yl)-1*H*-indole
R=for compound 1=H, 2=3-ClC$_6$H$_3$,4=2-ClC$_6$H$_3$, 5=C$_6$H$_5$

Yan Li etal[51] Synthesized fifteen novel (E)-a-(methoxyimino)-benzeneacetate derivatives.Bioassays indicated that compound 1a-1o showed potent fungicidal activity against Rhizoctoniasolani, Botrytis cinereapers, Gibberella zeae, Physalospora piricola and Bipolaris mayclis and 1a-1o showed potent fungicidal activity.



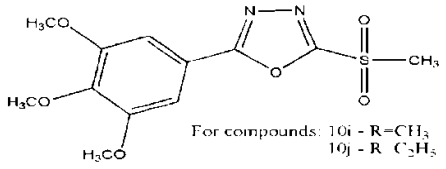
Compound 1a-1o

1a:R=H
1b:R=4-OMe
1c:R=4-C$_6$H$_5$ClH$_2$O
1d:R=3-Cl
1e:R=2,3-Cl$_2$
1f:R=2,4-Cl$_2$
1g:R=2,5Cl$_2$
1h:R=2,4-Cl$_2$,5F
1i:R=2F
1j:R=4F
1k:R=2F,4-Br
1l:R=2,3F$_2$
1m:R=4CF$_3$
1n:R=2-I
1o:R=4-NO$_2$

Fig: (E)-methyl 2-(methoxyimino)-2-(2-((5-phenyl-1, 3, 4-oxadiazol-2-ylthio) methyl)phenyl)acetate

Bao-An Song etal[52] Synthesized compounds using the key intermediates 5-(3,4,5-trimethoxyphenyl)-1,3,4-thiadiazole-2-thiol or the oxadiazole analogue and tested for Fungicidal activity. From all the synthesized compounds 10i and 10j can inhibit mycelium growth by approximately 50% *in vitro* against ten kinds of Fungus

.



For compounds: 10i - R=CH$_3$
10j - R C$_2$H$_5$

Fig: 2-(methylsulfonyl)-5-(3, 4, 5-trimethoxyphenyl)-1,3,4-oxadiazole derivatives

Wildersmith AE etal[53] Synthesized and found Tuberculostatic and Leprostatic properties in a series of 5-(4-pyridyl)-1, 3, 4- oxadiazole-2-thione, and 5-(4-pyridyl)-1,3,4-oxadiazole-2-one were tested for their Anti TB activity against Mycobacterium and found that all synthesized compound were active.

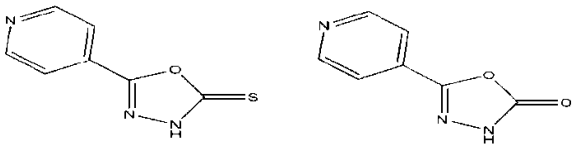


Fig: 5-(4-pyridyl)-1, 3, 4- oxadiazole-2-thione, 5-(4-pyridyl)-1, 3, 4-oxadiazole-2-one derivatives

Mir I etal[54] synthesized alpha [5-(2-furyl)-1,3,4-oxadiazol-2-yl-thio] acetohydrazine and tested for their activity. All compounds showed in vitro activity against Mycobacterium tuberculosis.

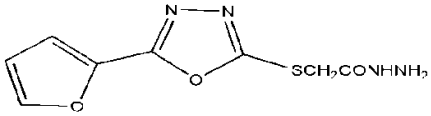


Fig: alpha [5-(2-furyl)-1, 3, 4-oxadiazol-2-yl-thio] acetohydrazine derivatives

Kapoor etal[55] Synthesized5-aryl-2- [N-(5-nitrofurfurylidene)] and 5-aryl-2-(N-thiocarbomylamino)-1,3,4-oxadiazole derivatives and tested for Antimicrobial activity all compound reported in were active.

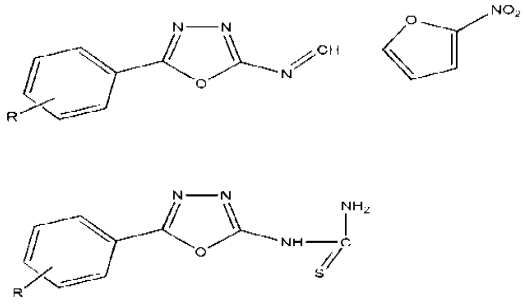
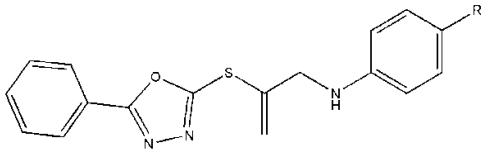


Fig: aryl-2- [N-(5-nitrofurfurylidene)] and 5-aryl-2-(N-thiocarbomylamino)-1, 3, 4-oxadiazole

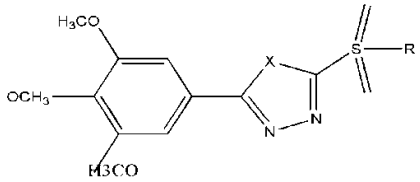
Oxadiazole as Antibacterial

Nilesh H. Patel etal[56] synthesised in vitro antibacterial activity of new oxoethylthio-1,3,4-oxadiazole derivatives. Newly synthesized compounds were tested for their in vitro anti-tubercular activity against Mycobacterium tuberculosis H37Rv using the BACTEC 460 radiometric system



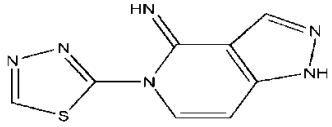
Oxadiazole as Antifungal

Bao-An Song etal[57] synthesized antifungal activities of 5-(3,4,5-trimethoxyphenyl)-2-sulfonyl-1,3,4-oxadiazole derivatives. The compounds have been shown to be fungicidally active. Title compounds 10i and 10j can inhibit mycelia growth by approximately 50% (EC50) at 2.9–93.3 lg/mL in vitro against fungi.

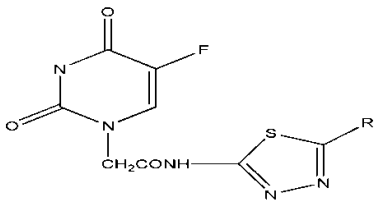


Thiadiazole as Antitumour

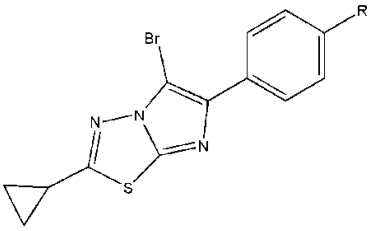
Xin Jian Song etal[58] synthesized some novel fluorinated pyrazolo[3,4-d]pyrimidine derivatives containing 1,3,4-thiadiazole as potential antitumor agents.



Kai Bo Zheng etal[59] synthesized of N1-acetylamino-(5-alkyl/aryl-1,3,4-thiadiazole-2-yl)-5-fluorouracil derivatives were designed and synthesized. The results of antitumor inhibitory activity test showed that some compounds possess more potent antitumor inhibitory activity than 5-fluorouracil.



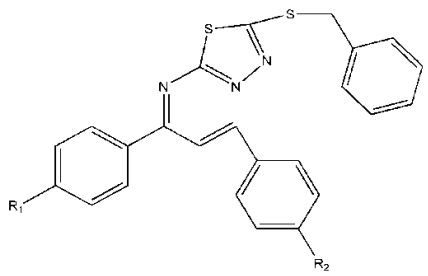
Andanappa.k.Gadad etal[60] synthesized anticancer evaluation of novel 2-cyclopropylimidazo[2,1-b] [1,3,4]-thiadiazole derivatives. the compounds tested, 5-bromo-6-(4-chlorophenyl)-2-cyclopropylimidazo[2,1-b][1,3,4]thiadiazole 5b was found to be the most active candidate of the series at five dose level screening with degree of selectivity toward Leukemic cancer cell line.



Where R=5b=4-Cl

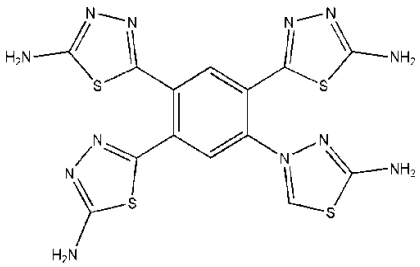
Thiadiazole as Antidepressant

Mohammad Yusuf, etal[61] synthesized anti-depressant activity of 5-amino-1, 3, 4-thiadiazole-2-thiol imines and thiobenzyl derivatives. 5-{[1-(4-chlorophenyl)-3-(4-methoxy-phenyl)prop-2-en-1-ylidene]-amino}- 5-benzylthio-1, 3,4 –thiadiazole 4i(b) and 5-{[1-(4-chlorophenyl)-3-(4-dimethyl-aminophenyl)-prop-2-en-1-ylidene]amino}-5- benzylthio-1,3,4-thiadiazole 4i(c) have shown significant anti-depressant activity, All the compounds in the series have passed neurotoxicity tests.



Where R1=H,OCH3,(CH3)2N,Cl,OH;R2=H,Cl

Nadia Salih etal [62] have been reported antimicrobial screening of tetra Schiff bases of 1,2,4,5-tetra (5-amino-1,3,4-thiadiazole-2-yl)benzene.The synthesized compounds 1,2,4,5-tetra[5-(4-nitrobenzylideneamino)-1,3,4-thiadiazole- 2yl]benzene was found to be the most potent antimicrobial activity.



OBJECTIVE

OBJECTIVE

We report the new synthesis of 1,3,4-oxadiazoles comprising 1,3,4- thiadiazole derivatives. The chemistry and Pharmacology of oxadiazole and thiadiazole have been of great interest because, of its various biological activities, so that the biological and pharmacological activity of oxadiazoles and thiadiazoles may be taken in to account for synergism.

It is well known that the introduction of mannich base in to an organic molecules causes dramatic changes in its biological profile. Therefore it was thought worth while to synthesize better kinds of drugs by incorporating 1,3,4- oxadiazole and 1,3,4-thiadiazole moiety.

In search for new bioactive potent molecule, it was thought worth while to incorporate some additional heterocyclic moieties in the oxadiazole and thiadiazole nucleus and study their biological and pharmacological activity, the review of literature reveal prompted us to synthesize substituted 1,3,4-oxadiazole and 1,3,4-thiadiazole compounds and those will be screened for antimicrobial, anti-inflammatory, anticancer and antitubercular activity to get potent bioactive molecule.

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PLAN OF WORK

1. Synthesis of 1, 3, 4- oxadiazole and 1,3,4-thiadiazole derivatives

Step-I:Synthesis of semicarbazone and thiosemicarbazone

Synthesis of titled compound was started from substituted aromatic aldehyde, which upon reaction with semicarbazides and thiosemicarbazide in alcohol to form semicarbazone and thiosemicarbazone respectively.

Step-II :Synthesis of 2-amino5-substituted 1,3,4-oxadiazole and 1,3,4-thiadiazole

Semicarbazone and thiosemicarbazone was suspended in warm water; ferric chloride in warm water was added quantitatively, slowly with constant stirring. The contents were heated at 100°C for 2hrs. Solution was filtered and citric acid , sodium carbonate were added. The obtained mixture was divided in to four portions and each portion was neutralized with ammonia respectively. The precipitate obtained was recrystallized from alcohol.

Step-III:Synthesis of Mannich base

The 2-amino-5-substituted-1, 3, 4-oxadiazole and 2-amino-5-substituted-1,3,4-thiadiazole were undergoes mannich reaction with substituted aniline and formaldehyde to get mannich base 2-amino 5-substituted 1, 3, 4- oxadiazole and 2-amino 5-substituted 1, 3, 4- thiadiazole derivatives respectively.

2. **Characterization**

The synthesized compounds were characterized by **UV, IR, NMR,** and **Mass** spectrum.

3. Biological activity

The synthesized compound will be evaluated for biological activity such as

a. Antibacterial
b. Antifungal
c. Anticancer
d. Antitubercular

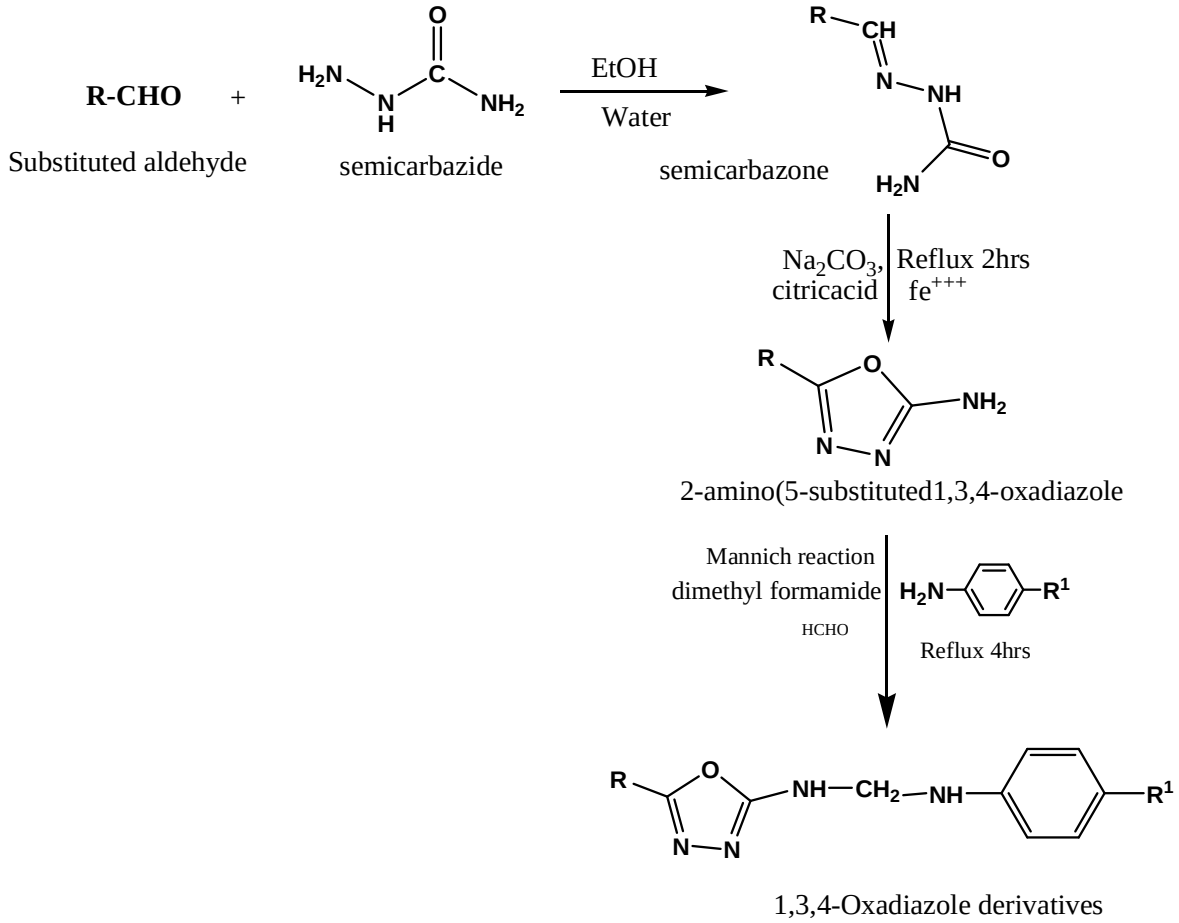
EXPERIMENTAL WORK

Experimental work

Synthetic Work

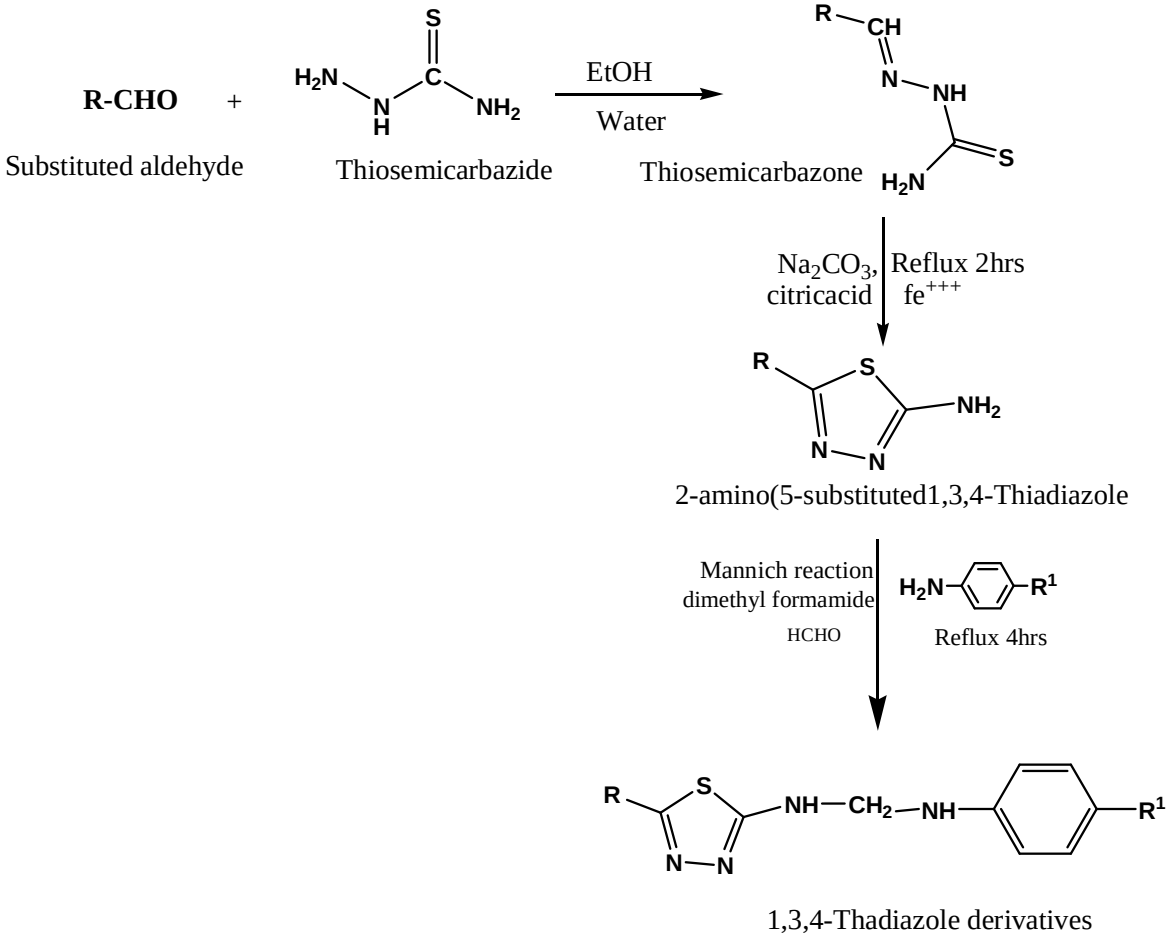
Scheme-I

Synthesis of 1,3,4- Oxadiazole derivatives



Scheme-II

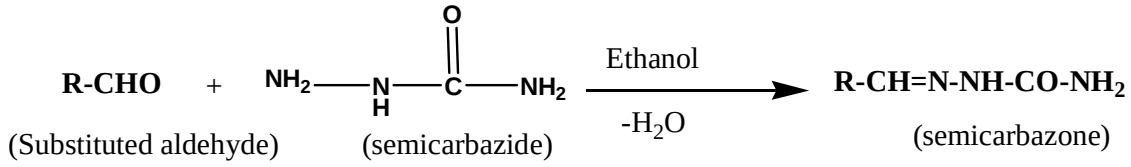
Synthesis of 1, 3, 4 -Thiadiazole derivatives



I. Synthesis of 1,3,4-oxadiazole derivatives

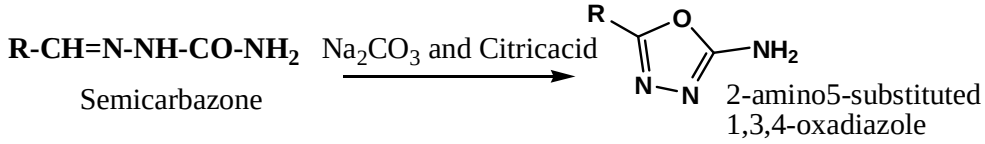
Step-I: Synthesis of semicarbazone

Synthesis of titled compound was started from substituted aromatic aldehyde, which upon reaction with semicarbazides in alcohol to form semicarbazone..



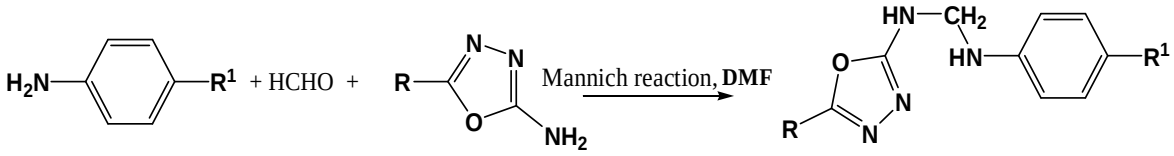
Step-II: Synthesis of 2-amino 5-substituted 1,3,4- oxadiazole

Semicarbazone (0.01M) was suspended in 300ml of warm water; ferric chloride (0.01M) in 300ml of warm water was added quantitatively, slowly with constant stirring. The contents were heated at 100°C for 2hrs. Solution was filtered and citric acid (0.11M), sodium carbonate (0.05M) were added. The obtained mixture was divided in to four portions and each portion was neutralized with ammonia. The precipitate obtained was recrystallized from alcohol.



Step-III: Synthesis of mannich base

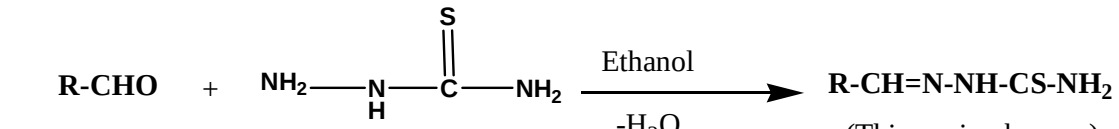
The 2-amino-5-substituted-1, 3, 4-oxadiazole were undergoes mannich reaction with substituted aniline and formaldehyde to get mannich base 2-amino 5-substituted 1,3,4-oxadiazole derivatives.



I Synthesis of 1,3,4-thiadiazole derivatives

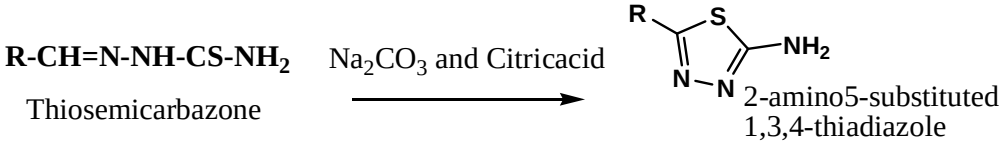
Synthesis of titled compound was started from substituted aromatic aldehyde, which upon reaction with thiosemicarbazides in alcohol to form thiosemicarbazone.

Step-I: Synthesis of thiosemicarbazone



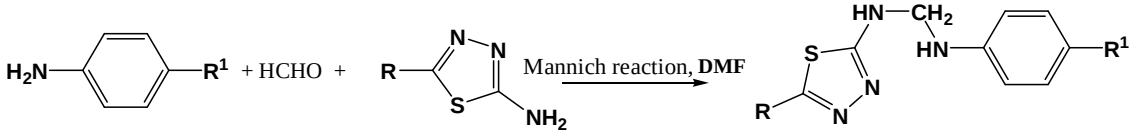
Step-II: Synthesis of 2-amino 5-substituted 1,3,4-Thiadiazole

Thiosemicarbazone (0.01M) was suspended in 300ml of warm water; ferric chlorid (0.01M).In 300ml of warm water was added quantitatively, slowly with constant stirring. The contents were heated at 100°C for 2hrs. Solution was filtered and citric acid (0.11M), sodiumcarbonate (0.05M) were added. The obtained mixture was divided in to four portions and eachportion was neutralized with ammonia. The precipitate obtained was recrystallized from alcohol



Step-III: Synthesis of Mannich base

The 2-amino-5-substituted1,3,4-thiadiazole were undergoes mannich reaction with substituted aniline and formaldehyde to get mannich base 2-amino5-substituted1,3,4-thiadiazole derivatives.



II. Characterization

Characterization of the synthesized compounds by the analytical technique like

- **Thin layer chromatography**
- **Infrared spectral analysis**
- **Nuclear magnetic resonance spectral analysis**
- **Mass Spectroscopy**

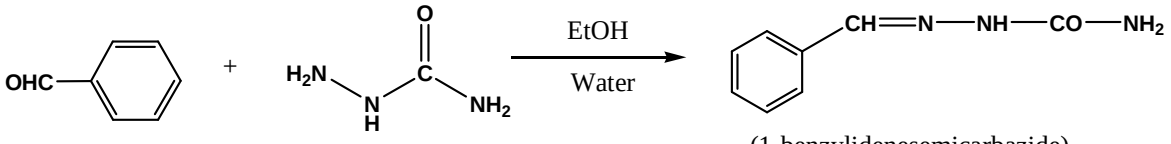
III.Biological Activity

Screening of the synthesized compounds for **antibacterial** , **antifungal** activity, **anticancer** and **antitubercular** activity.

Procedure for the synthesis of individual compound

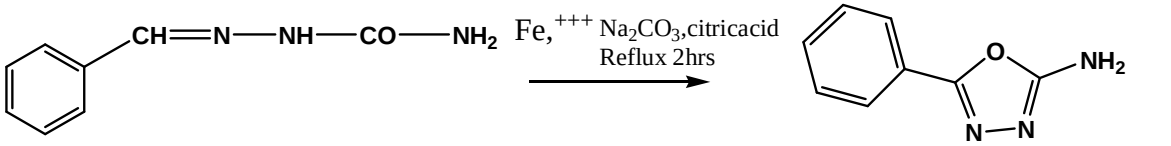
Synthesis of compound 1B

Step-I



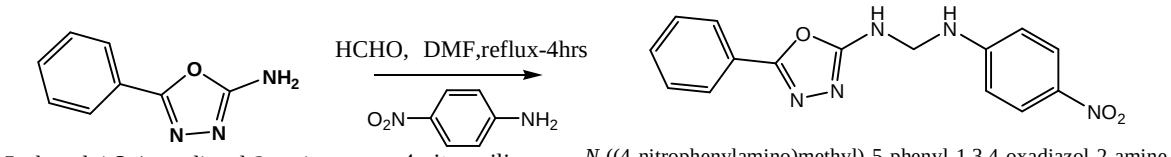
(Benzaldehyde) (semicarbazide) (1-benzylidenesemicarbazide)

StepII



1-benzylidenesemicarbazide 5-phenyl-1,3,4-oxadiazol-2-amine

Step-III



5-phenyl-1,3,4-oxadiazol-2-amine 4-nitroaniline *N*-((4-nitrophenylamino)methyl)-5-phenyl-1,3,4-oxadiazol-2-amine

Procedure

To 0.01mole(0.75gm) of semicarbazide in 10ml of water added 0.01 mole (1.06gm) of benzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated 1-benzylidene semicarbazide was filtered and dried. To 0.05 mole (8.15gm) of 1-benzylidene semicarbazide in 300ml warm water, 0.01 mole (1.62gm) of ferric chloride in 300ml water was added slowly with stirring .The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1mole (19.2gm) of citricacid and 0.05 mole (5.25gm) of sodium carbonate.The resulting mixture was neutralised with four portions

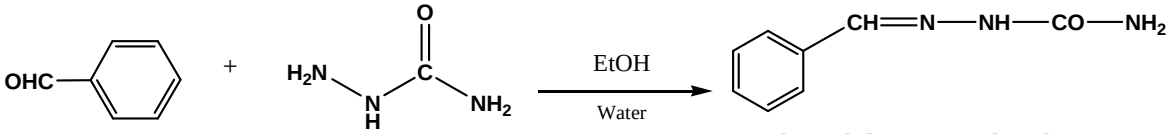
of ammonia. The precipitated 5-phenyl1,3,4-oxadiazol-2amine was seperated by filteration and recrystallised from ethanol.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde (37-48%) and 0.02 mole (2.76gm) of 4-nitroaniline, drop by drop with stirring.

The resulting mixture was stirred well and refluxed for 4-5hrs, poured in to the cold water. The precipitated 0.025mole (7.16gm) of N-((4-Nitrophenylamino)methyl)-5-phenyl-1,3,4-oxadiazol-2-amine was filtered and dried.The product obtained was recrystallised from ethanol.

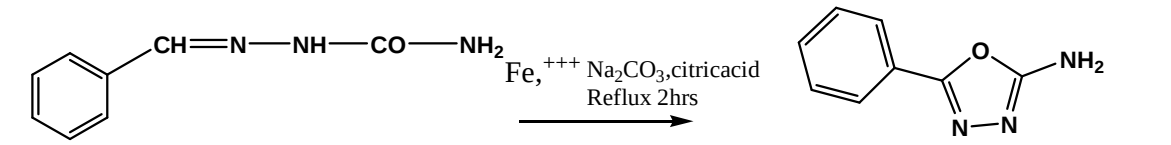
Synthesis of compound 1E:4-({((5-phenyl-1,3,4-oxadiazol-2-yl)amino]methyl}amino)benzene sulphonamide

Step-I



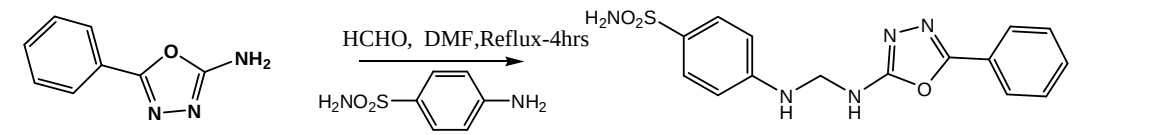
Benzaldehyde semicarbazide 1-benzylidenesemicarbazide

Step-II



1-benzylidenesemicarbazide 5-phenyl-1,3,4-oxadiazol-2-amine

Step-III



5-phenyl-1,3,4-oxadiazol-2-amine sulphanilamide 4-({[(5-phenyl-1,3,4-oxadiazol-2-yl)amino]methyl}amino)benzenesulfonamide

Procedure

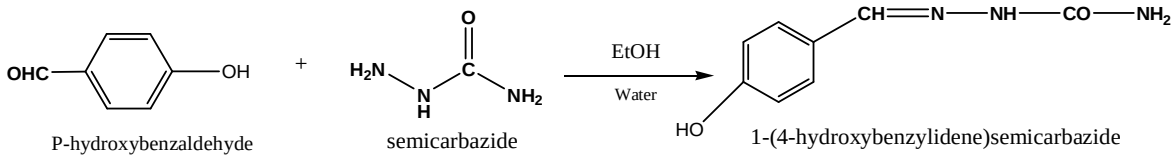
Dissolve 0.01mole(0.75gm) of semicarbazide in 10ml of warm water added 0.01 mole(1.06gm) of benzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated 1-benzylidene semicarbazide was filtered and dried.To 0.05 mole (8.15gm) of 1-benzylidene semicarbazide in 300ml warmwater, 0.01mole(1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole (19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with four portions of ammonia. The precipitated 5-phenyl1,3,4-oxadiazol-2amine was seperated by filteration and recrystallised from ethanol.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde (37-48%) and 0.02 mole (3.44gm) of sulphanilamide drop by drop with stirring.

The resulting mixture was stirred well and refluxed for 4-5hrs, poured intocoldwater.Theprecipitated0.025mole(7.45gm)of4({[(5phenyl1,3,4oxadiazolyl)amino]methyl}amino)benzene sulphonamide was filtered and dried. The product obtained was recrystallised from ethanol.

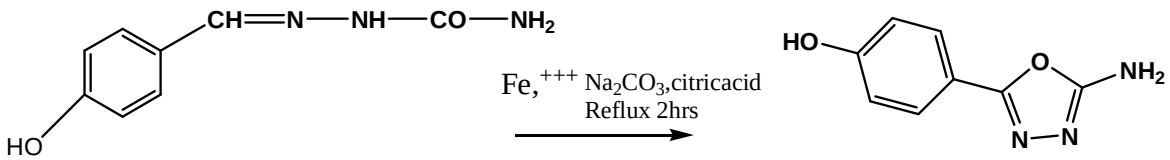
Synthesis of compound 2B:N-((4-nitrophenylamino)methyl)-5-phenyl-1,3,4-oxadiazol-2-amine

Step-I



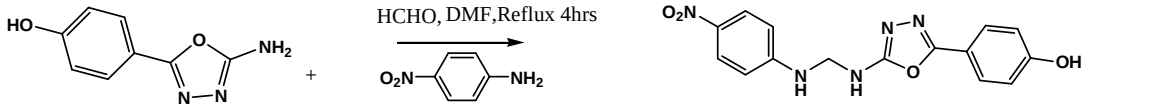
P-hydroxybenzaldehyde semicarbazide 1-(4-hydroxybenzylidene)semicarbazide

Step-II



1-(4-hydroxybenzylidene)semicarbazide 4-(5-amino-1,3,4-oxadiazol-2-yl)phenol

Step-III



4-(5-amino-1,3,4-oxadiazol-2-yl)phenol 4-nitroaniline 4-(5-((4-nitrophenylamino)methylamino)-1,3,4-oxadiazol-2-yl)phenol

Procedure

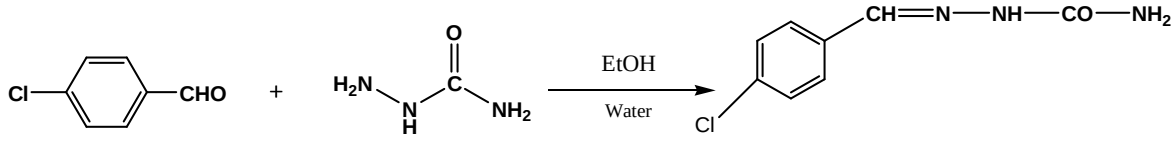
Dissolve 0.01mole (0.75gm) of semicarbazide in 10ml of warm water and 0.01 mole (1.22gm)of P-hydroxybenzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated1-(4-hydroxybenzylidene)semicarbazide was filtered and dried. To 0.005 mole(0.89gm) of 1--(4-hydroxybenzylidene) semicarbazide in 300ml warmwater, 0.01mole (1.62gm) of ferric chloride in 300ml water was added slowly with stirring.The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.21gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with four portions of ammonia. The precipitated 4-(5-amino-1,3,4-oxadiazol-2-yl) phenol was seperated by filteration and recrystallised from ethanol.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde (37-48%) and 0.02 mole (2.76gm) of p-nitroaniline drop by drop with stirring.

The resulting mixture was stirred well and refluxed for 4-5hrs, poured tocoldwater.Theprecipitated0.025mole(7.86gm)of4(5(4nitrophenylamino]methyl}amino)1,3, 4oxadiazol-2-yl)phenol was filtered and dried. The product obtained was recrystallised from ethanol.

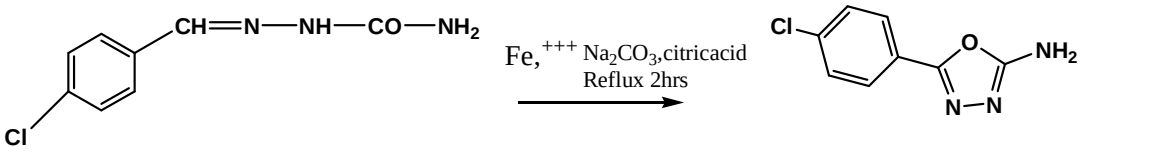
Synthesis of compound 2C:N-((4-methoxyphenylamino)methyl)-5-(4-chlorophenyl)-1,3,4-oxadiazol-2-amine

Step-I



Parachlorobenzaldehyde 1-(4-chlorobenzylidene)semicarbazide

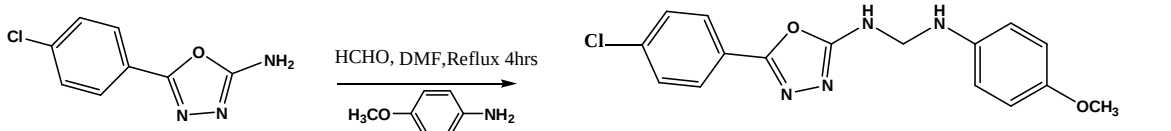
Step-II



1-(4-chlorobenzylidene)semicarbazide

4-(5-amino-1,3,4-oxadiazol-2-yl)chlorophenol

Step-III



4-(5-amino-1,3,4-oxadiazol-2-yl)chlorophenol

4-methoxyaniline

N-((4-methoxyphenylamino)methyl)-5-(4-chlorophenyl)-1,3,4-oxadiazol-2-amine

Procedure

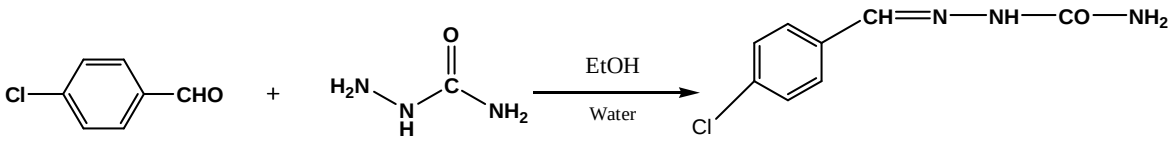
Dissolve 0.01mole (0.75gm)of semicarbazide in 10ml of warm water and 0.01 mole (1.40gm)of P-chlorobenzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated 1-(4-chlorobenzylidene)semicarbazide was filtered and dried.To 0.5 mole(0.98gm) of 1-(4-chlorobenzylidene)semicarbazide in 300ml warmwater, 0.01mole(1.62gm) of Ferric chloride in 300ml water was added slowly with stirring.The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole(5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with four portions of ammonia. The precipitated 4-(5-amino-1,3,4-oxadiazol-2-yl) chloro phenol was seperated by filteration and recrystallised from ethanol.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde (37-48%) and 0.02 mole (2.46gm) of p-methoxyaniline drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the cold water. The precipitated 0.025mol(7.86gm) of N-((4-methoxyphenylamino]methyl)-5-(4-Chlorophenyl)- 1,3,4-oxadiazol-2-amine was filtered and dried. The product obtained was recrystallised from ethanol.

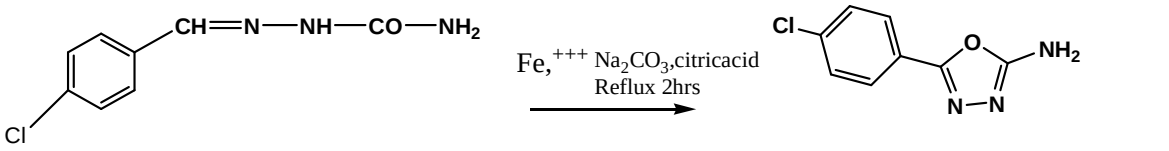
Synthesis of compound 3B:N-[5-(4-Chlorophenyl)-1,3,4-oxadiazol-2-yl]-N^1-(4-nitrophenyl)methanediamine

Step-I



Parachlorobenzaldehyde semicarbazide 1-(4-chlorobenzylidene)semicarbazide

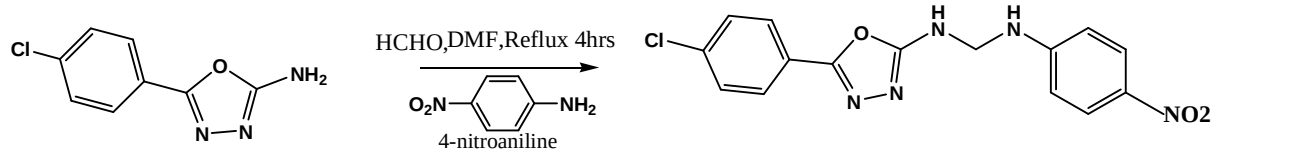
Step-II



1-(4-chlorobenzylidene)semicarbazide 4-(5-amino-1,3,4-oxadiazol-2-yl)chlorophenol

Step-III

.



4-(5-amino-1,3,4-oxadiazol-2-yl)chlorophenol 4-nitroaniline *N*-[5-(4-chlorophenyl)-1,3,4-oxadiazol-2-yl]-*N'*-(4-nitrophenyl)methanediamine

Procedure

Dissolve 0.01mole (0.75gm)of semicarbazide in 10ml of warm water added 0.01 mole(1.22gm) of P-hydroxybenzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated 1-(4-chlorobenzylidene)semicarbazide was filtered and dried.To 0.5mole(0.98gm) of 1-(4-chlorobenzylidene)semicarbazide in 300ml warmwater, 0.01mole(1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole(5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with four portions of ammonia. The precipitated 4-(5-amino-

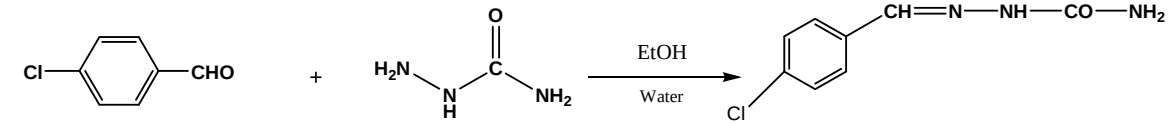
1,3,4- oxadiazol-2-yl)chlorophenol was seperated by filteration and recrystallised from ethanol.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde (37-48%) and 0.02 mole (2.76gm) of p-nitroaniline drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the cold water. The precipitate 0.025mole (8.7gm) of N-(5-(4-Chlorophenyl)-1,3,4-oxadiazol-2-yl]-N^1–(4-nitrophenyl)methanediamine was filtered and dried. The product obtained was recrystallised from ethanol.

Synthesis of compound 3C:N-((4-methoxyphenylamino)methyl)-5-(4-chlorophenyl)-1,3,4-oxadiazol-2-amine

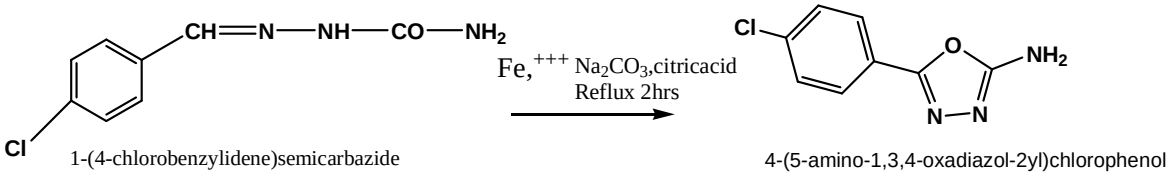
Step-I



Parachlorobenzaldehyde

1-(4-chlorobenzylidene)semicarbazide

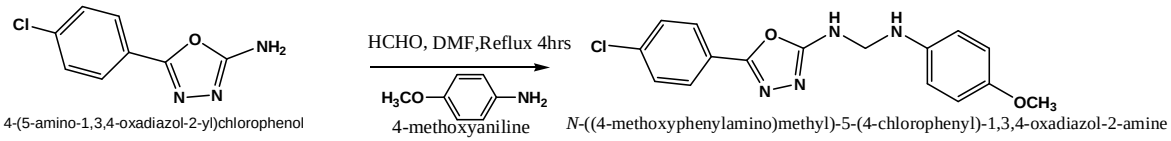
Step-II



1-(4-chlorobenzylidene)semicarbazide

4-(5-amino-1,3,4-oxadiazol-2yl)chlorophenol

Step-III



4-(5-amino-1,3,4-oxadiazol-2-yl)chlorophenol

4-methoxyaniline

N-((4-methoxyphenylamino)methyl)-5-(4-chlorophenyl)-1,3,4-oxadiazol-2-amine

Procedure

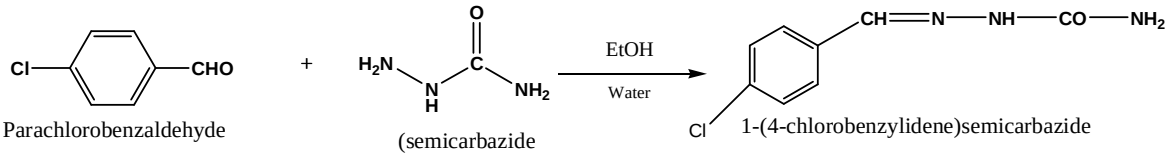
Dissolve 0.01mole (0.75gm) of semicarbazide in 10ml of warm water added 0.01 mole(1.40gm) of P-chlorobenzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated 1-(4-chlorobenzylidene)semicarbazide was seperated by filteration.To 0.005mole (0.98gm)1-(4-chlorobenzylidene)semicarbazide in 300ml warmwater, 0.01mole (1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with four portions of ammonia. The precipitated 4-(5-amino-1,3,4-oxadiazol-2-yl)chlorophenol was seperated by filteration and recrystallised from ethanol.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (2.46gm) of p-methoxyaniline drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the coldwater.The precipitate 0.025mole(8.50gm) of N((4Methoxyphenylamino)methyl) 5(4chlorophyl-1,3,4-oxadiazol-2-yl)chlorophenol was filtered and dried. The product obtained was recrystallised from ethanol.

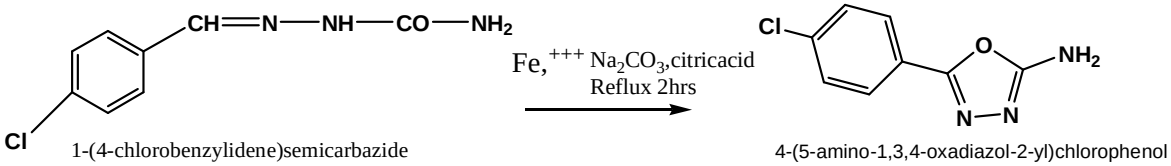
Synthesis of compound 3D:N-(4-Chlorophenyl)-N^1-[5-(4-Chlorophenyl)-1,3,4-oxadiazol-2-yl]methanediamine

Step-I



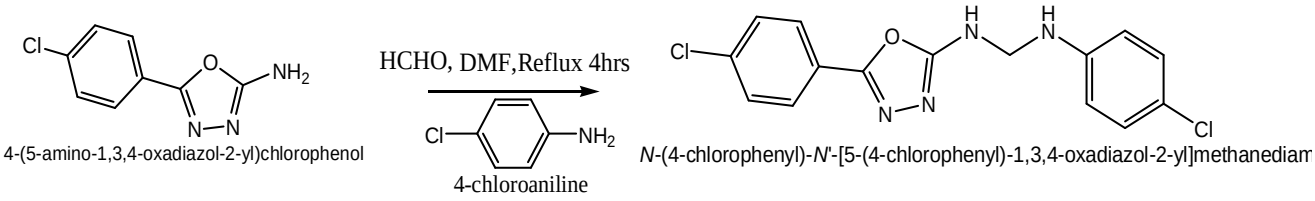
Parachlorobenzaldehyde (semicarbazide 1-(4-chlorobenzylidene)semicarbazide

Step-II



1-(4-chlorobenzylidene)semicarbazide 4-(5-amino-1,3,4-oxadiazol-2-yl)chlorophenol

Step-III



4-(5-amino-1,3,4-oxadiazol-2-yl)chlorophenol

4-chloroaniline

N-(4-chlorophenyl)-*N*'-[5-(4-chlorophenyl)-1,3,4-oxadiazol-2-yl]methanediam

Procedure

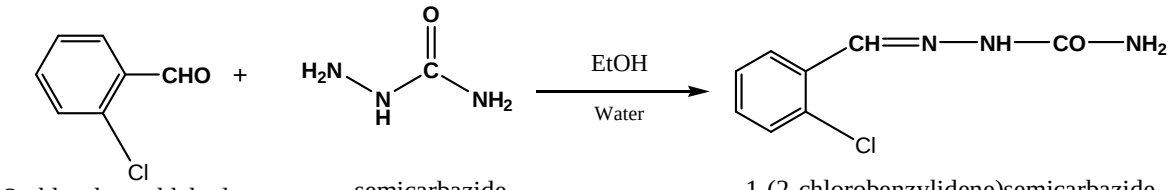
Dissolve 0.01mole (0.75gm)of semicarbazide in 10ml of warm water and 0.01 mole (1.40gm)of P-chlorobenzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated 1-(4-chlorobenzylidene)semicarbazide was filtered and dried. To 0.005mole (0.98gm) of 1-(4-chlorobenzylidene)semicarbazide in 300ml warm water, 0.01mole(1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with four portions of ammonia. The precipitated 4-(5-amino-1,3,4-oxadiazol-2-yl)chlorophenol was seperated by filteration and recrystallised from ethanol.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (2.55gm) of p-Chloroaniline drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the cold water. The precipitate 0.025mole(9.16gm) of N-((4-Chlorophenyl)N^1-[5-(4-chlorophenyl)-1,3,4-oxadiazol-2-yl)methanediamine was filtered and dried.The product obtained was recrystallised from ethanol.

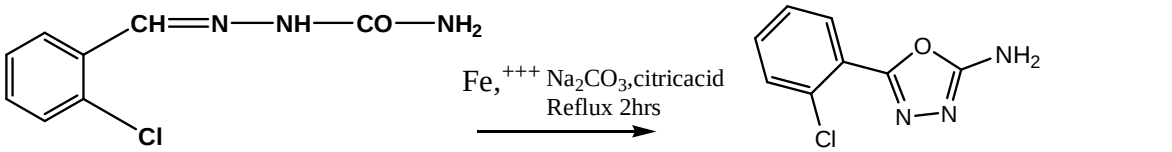
Synthesis of compound 4B:N-[5-(2-Chlorophenyl)-1,3,4-oxadiazol-2-yl]-N^1-(4-nitrophenyl)methanediamine

Step-I



2-chlorobenzaldehyde semicarbazide 1-(2-chlorobenzylidene)semicarbazide

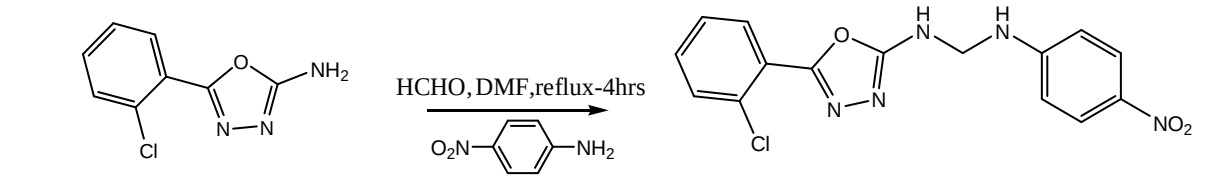
Step-II



1-(2-chlorobenzylidene)semicarbazide

5-(2-chlorophenyl)-1,3,4-oxadiazol-2-amine

Step-III



(5-(2-chlorophenyl)-1,3,4-oxadiazol-2-amine)) (nitroaniline) *N*-[5-(2-chlorophenyl)-1,3,4-oxadiazol-2-yl]-*N*'-(4-nitrophenyl)methanediamine

Procedure

Dissolve 0.01mole (0.75gm) of semicarbazide in 10ml of warm water added 0.01 mole of 2-chlorobenzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated 1-(2-chlorobenzylidene)semicarbazide was filtered and dried.To 0.005 mole(0.96gm) of 1-(2-chlorobenzylidene)semicarbazide in 300ml warm water, 0.01mole(1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole(5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with four portions of ammonia. The precipitated (5-(2-

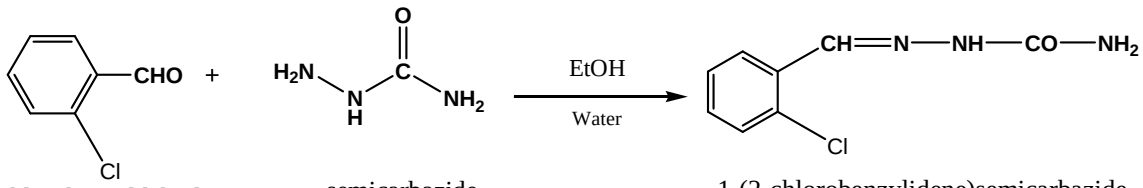
chlorophenyl)- 1,3,4-oxadiazol-2-amine was seperated by filteration and recrystallised from ethanol.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (2.76gm) of p-nitroaniline drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the cold water. The precipitate 0.025mole (7.89gm) of N[5chlorophenyl)-1,3,4-oxadiazol-2-yl]-N(4nitrophenyl)methanediamine was filtered and dried. The product obtained was recrystallised from ethanol.

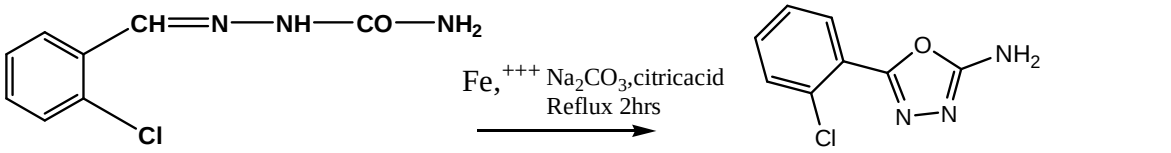
Synthesis of compound 4E:4-[({[5-(2-Chlorophenyl)-1,3,4-oxadiazol-2-yl]amino}methyl)amino]benzenesulphonamide

Step-I



2-chlorobenzaldehyde semicarbazide 1-(2-chlorobenzylidene)semicarbazide

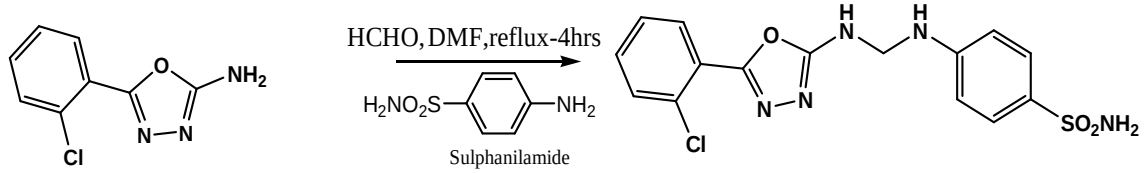
Step-II



1-(2-chlorobenzylidene)semicarbazide

5-(2-chlorophenyl)-1,3,4-oxadiazol-2-amine

Step-III



5-(2-chlorophenyl)-1,3,4-oxadiazol-2-amine

4-[({[5-(2-chlorophenyl)-1,3,4-oxadiazol-2-yl]amino}methyl)amino]benzenesulfonami

Procedure

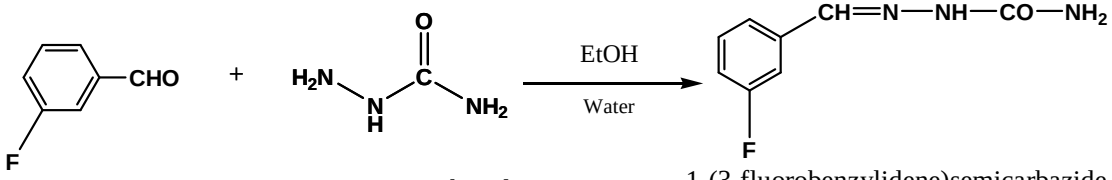
Dissolve 0.01mole (0.75gm) of semicarbazide in 10ml of warm water added 0.01 mole of 2-chlorobenzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated 1-(2-chlorobenzylidene)semicarbazide was filtered and dried.To 0.005 mole(0.96gm) of 1-(2-chlorobenzylidene)semicarbazide in 300ml warm water, 0.01mole(1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with four portions of ammonia. The precipitated (5-(2-chlorophenyl)- 1,3,4-oxadiazol-2-amine was seperated by filteration and recrystallised from ethanol.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (3.44gm) of p-sulphanilamide drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the coldwater. The precipitate 0.025mole(7.8gm) 4-[({[5-(2-Chlorophenyl)-1,3,4-oxadiazol-2-yl]amino}methyl)amino]benzene sulphonamide was filtered and dried. The product obtained was recrystallised from ethanol.

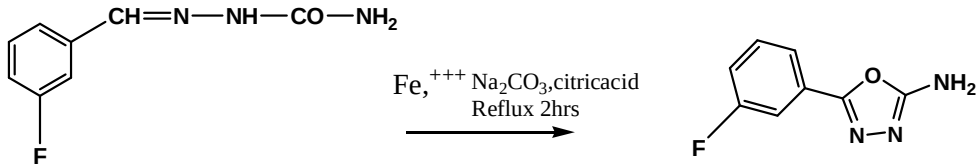
Synthesis of compound 5E: N-(4-Sulphonamyl)-N-[5-(3-Fluorophenyl)-1,3,4-oxadiazol-2-yl]methanediamine

Step1



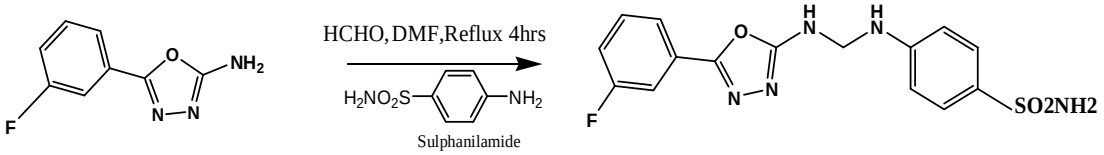
3-Fluorobenzaldehyde semicarbazide 1-(3-fluorobenzylidene)semicarbazide

StepII



1-(3-fluorobenzylidene)semicarbazide 5-(3-fluorophenyl)-1,3,4-oxadiazol-2-amine

Step-III



5-(3-fluorophenyl)-1,3,4-oxadiazol-2-amine

N-(4-Sulphonamyl)-N-[5-(3-Fluorophenyl)-1,3,4-oxadiazol-2-yl]methanediamine

Procedure

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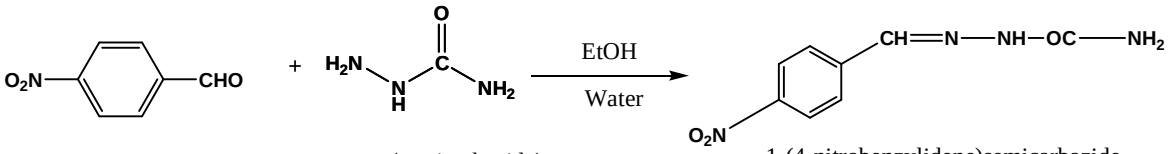
Dissolve 0.01mole (0.75gm) of semicarbazide in 10ml of warm water added 0.01 mole of 3-Flurobenzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated 1-(3-fluorobenzylidene)semicarbazide was filtered and dried. To 0.005 mole (0.90gm) of 1-(3-fluorobenzylidene)semicarbazide in 300ml warm water, 0.01mole (1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with four portions of ammonia. The precipitate(5-(3-fluorophenyl)-1,3,4-oxadiazol-2-amine) was seperated by filteration and recrystallised from ethanol.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (3.44gm) of p-Sulphanilamide drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the coldwater. The precipitate 0.025mole (7.8gm) of N-(4-Sulphonamyl)-N-[5-(3-Fluorophenyl)-1,3,4-oxadiazol-2-yl]methanediamine was filtered and dried. The product obtained was recrystallised from ethanol.

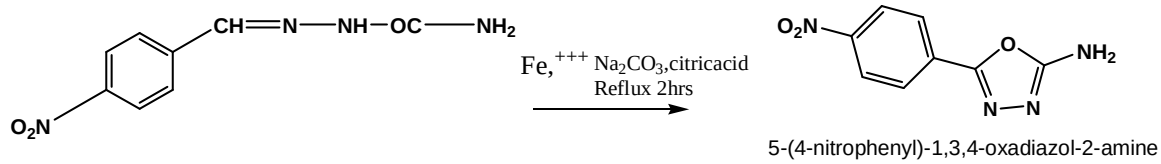
Synthesis of compound 6B:N-[5-(4-nitrophenyl)-1,3,4-oxadiazol-2-yl]-N^1-nitrophenylmethanediamine

StepI



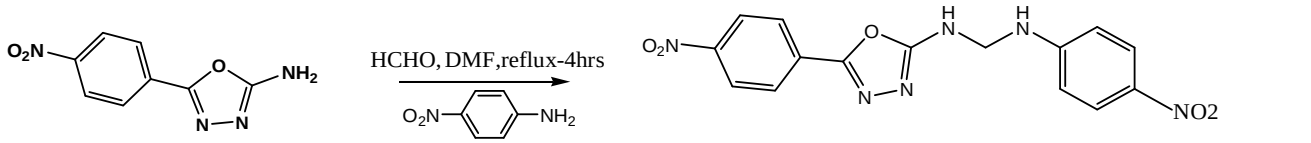
4-nitrobenzaldehyde (semicarbazide) 1-(4-nitrobenzylidene)semicarbazide

StepII



5-(4-nitrophenyl)-1,3,4-oxadiazol-2-amine

StepIII



5-(4-nitrophenyl)-1,3,4-oxadiazol-2-amine (nitroaniline)

N-[5-(4-nitrophenyl)-1,3,4-oxadiazol-2-yl]-N1-nitrophenylmethanedian

Procedure

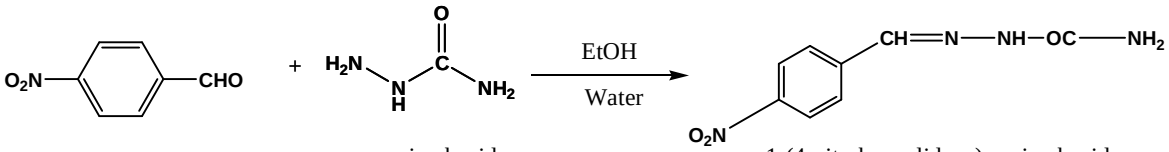
Dissolve 0.01mole (0.75gm) of semicarbazide in 10ml of warm water added 0.01 mole of 4-Nitrobenzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated 1-(4-Nitrobenzylidene)semicarbazide was filtered and dried To 0.005 mole (1.04gm) of 1-(4-Nitrobenzylidene)semicarbazide in 300ml warm water, 0.01mole (1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole(5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with four portions of ammonia. The precipitated(5-(4-Nitrophenyl)- 1,3,4-oxadiazol-2-amine) was seperated by filteration and recrystallised from ethanol.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (2.76gm) of p-nitroaniline drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the coldwater. The precipitate 0.025mole(7.75gm) of N-[5-(4-nitrophenyl)-1,3,4-oxadiazol-2-yl]-N1-nitrophenylmethanediamine was filtered and dried.The product obtained was recrystallised from ethanol.

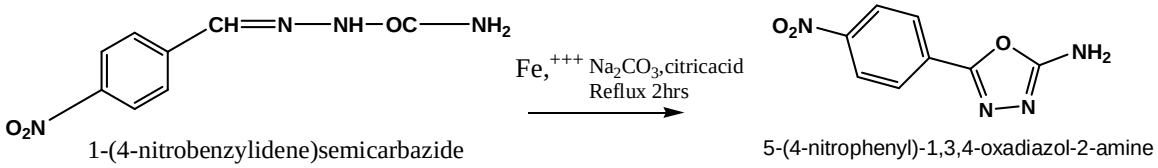
Synthesis of compound 6E: 4-[({[5-(4-nitrophenyl)-1,3,4-oxadiazol-2yl]amino}methyl)amino]benzenesulphonamide

StepI



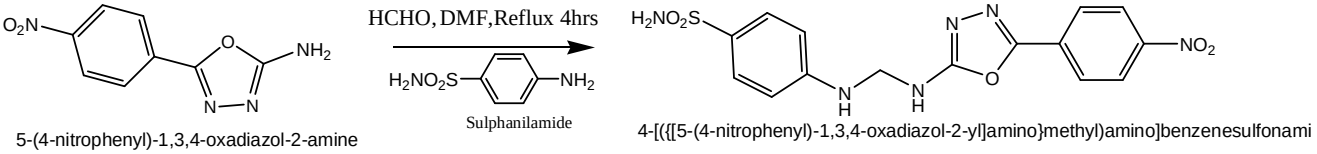
4-nitrobenzaldehyde semicarbazide 1-(4-nitrobenzylidene)semicarbazide

StepII



1-(4-nitrobenzylidene)semicarbazide 5-(4-nitrophenyl)-1,3,4-oxadiazol-2-amine

StepIII



5-(4-nitrophenyl)-1,3,4-oxadiazol-2-amine Sulphanilamide 4-[({[5-(4-nitrophenyl)-1,3,4-oxadiazol-2-yl]amino}methyl)amino]benzenesulfonami

Procedure

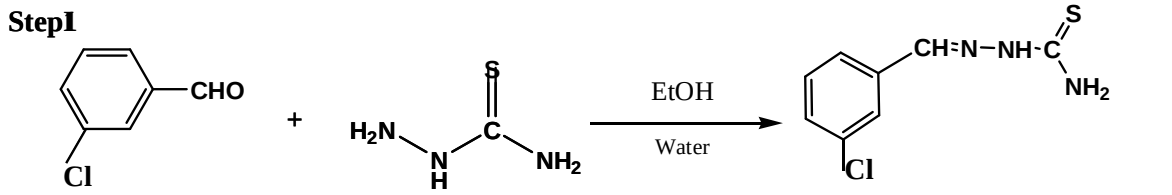
Dissolve 0.01mole (0.75gm) of semicarbazide in 10ml of warm water added 0.01 mole of 4-Nitrobenzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated 1-(4-Nitrobenzylidene)semicarbazide was filtered and dried. To 0.005 mole(1.04gm) of 1-(4-Nitrobenzylidene)semicarbazide in 300ml warm water, 0.01mole (1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole (19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised withfour portions of ammonia. The precipitate(5-(4-Nitrophenyl)- 1,3,4-oxadiazol-2-amine) was seperated by filteration and recrystallised from ethanol.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (3.44gm) of p-sulphanilamide drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the coldwater. The precipitate 0.025 mole(7.75gm) of 4-[({[5-(4-nitrophenyl)-1,3,4-oxadiazol-2yl]amino}methyl)amino]benzenesulphonamide was filtered and dried. The product obtained was recrystallised from ethanol.

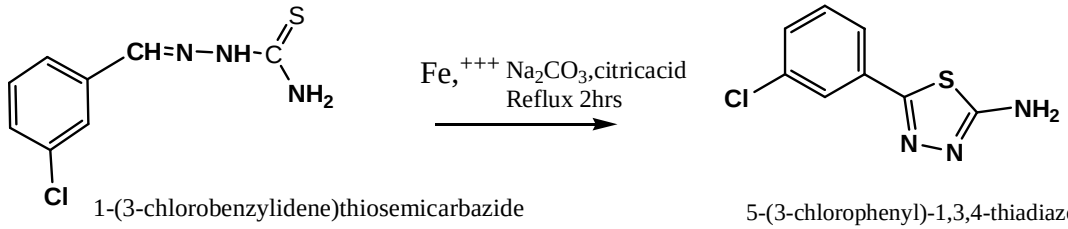
Synthesis of compound $8A_1$: 4-[({[5-(3-Chlorophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulphonamide

StepI



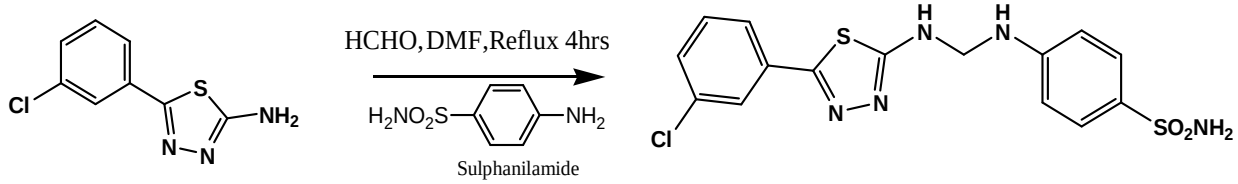
3-Chlorobenzaldehyde Thiosemicarbazide 1-(3-chlorobenzylidene)thiosemicarbazide

StepII



1-(3-chlorobenzylidene)thiosemicarbazide 5-(3-chlorophenyl)-1,3,4-thiadiazol-2-amine

StepIII



5-(3-chlorophenyl)-1,3,4-thiadiazol-2-amine 4-[({[5-(3-chlorophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulfonami

Procedure

Dissolve 0.01mole(0.91gm) of thio semicarbazide in 10ml of warm water added 0.01 mole(1.40gm) of 3-Chlorobenzaldehyde in 10ml of ethanol and stirred well

for 10mts, the precipitated 1-(3-Chlorobenzylidene) thiosemicarbazide was filtered and dried. To 0.005 mole (1.56gm) of 1-(3-Chlorobenzylidene)thiosemicarbazide in 300ml warm water, 0.01mole(1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with four portions of ammonia. The precipitated(5-(3-Chlorophenyl)- 1,3,4-thiadiazol-2-amine) was seperated by filteration and recrystallised from ethanol.

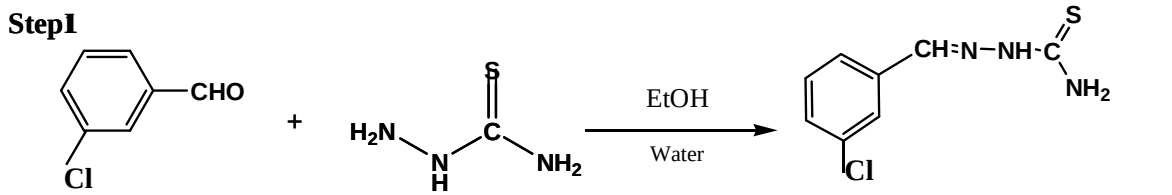
To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (3.45gm) of p-sulphanilamide drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the coldwater. The precipitate 0.025(9.85gm) of 4-[({[5-(3-Chlorophenyl)-1,3,4-thiadiazol-2yl]amino}methyl)amino]benzenesulphonamide was filtered and dried. The product obtained was recrystallised from ethanol.

Synthesis of compound $8A_2$:

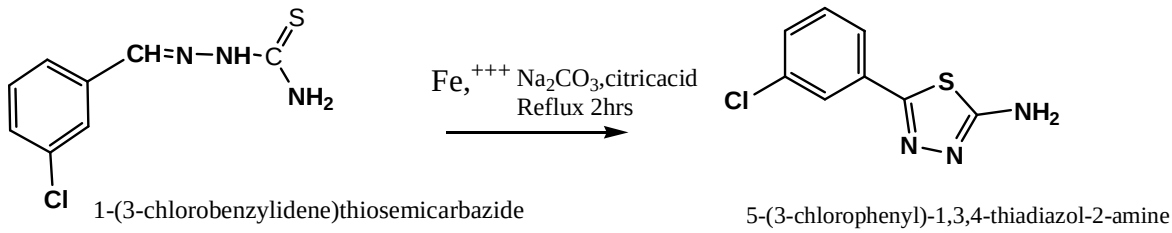
(4-Chlorophenyl)-N^1-[5-(3-Chlorophenyl)-1,3,4-thiadiazol-2-yl]methanediamine

StepI



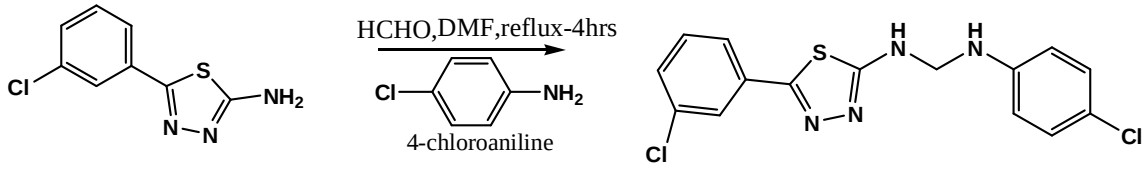
3-Chlorobenzaldehyde Thiosemicarbazide 1-(3-chlorobenzylidene)thiosemicarbazide

StepII



1-(3-chlorobenzylidene)thiosemicarbazide 5-(3-chlorophenyl)-1,3,4-thiadiazol-2-amine

StepIII



5-(3-chlorophenyl)-1,3,4-thiadiazol-2-amine

N-(4-chlorophenyl)-*N'*-[5-(3-chlorophenyl)-1,3,4-thiadiazol-2-yl]methanediamine

Procedure

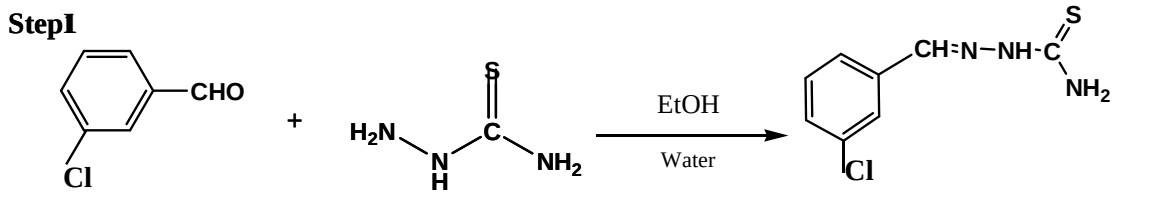
Dissolve 0.01mole(0.91gm) of thio semicarbazide in 10ml of warm water added 0.01 mole (1.40gm) of 3-Chlorobenzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated 1-(3-Chlorobenzylidene) thiosemicarbazide was filtered and dried.To 0.005mole (1.56gm) of 1-(3-Chlorobenzylidene)thiosemicarbazide in 300ml warm water, 0.01mole (1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole (19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with four portions of ammonia. The precipitated(5-(3-Chlorophenyl)- 1,3,4-thiadiazol-2-amine) was seperated by filteration and recrystallised from ethanol.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (2.55gm) of p-Chloroaniline drop by drop with stirring.

The reaction mixture was . stirred well and refluxed for 4-5hrs and poured in to the coldwater. The precipitate 0.025 mole(8.95gm) N-(4-Chlorophenyl)-N-[5-(3-Chlorophenyl)-1,3,4-thiadiazol-2yl]methanediamine was filtered and dried. The product obtained was recrystallised from ethanol.

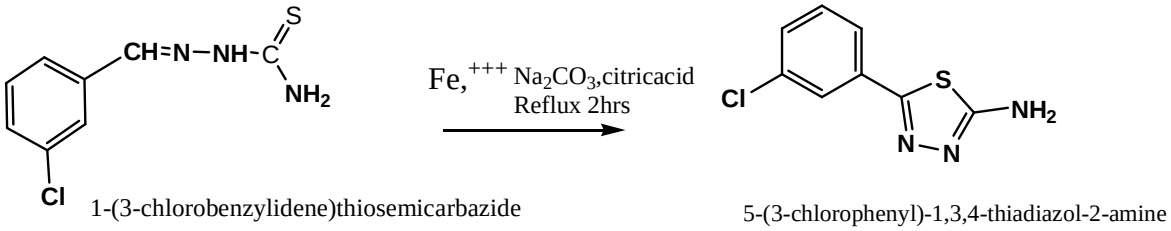
Synthesis of compound $8A_3$:N-[5-(3-Chlorophenyl)-1,3,4-thiadiazol-2-yl]-N^1-(4-nitrophenyl)methanediamine

StepI



3-Chlorobenzaldehyde Thiosemicarbazide 1-(3-chlorobenzylidene)thiosemicarbazide

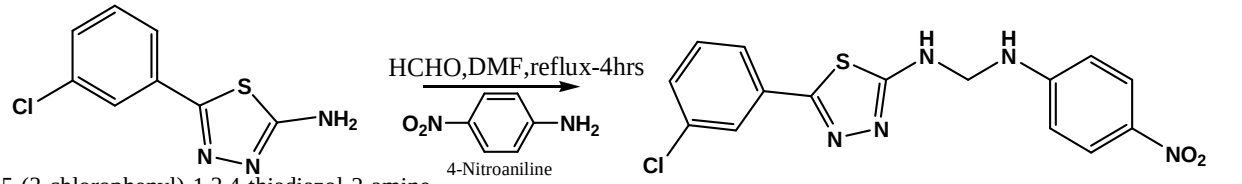
StepII



1-(3-chlorobenzylidene)thiosemicarbazide

5-(3-chlorophenyl)-1,3,4-thiadiazol-2-amine

StepIII



5-(3-chlorophenyl)-1,3,4-thiadiazol-2-amine

4-Nitroaniline

N-[5-(3-chlorophenyl)-1,3,4-thiadiazol-2-yl]- *N*'-(4-nitrophenyl)methanediamine

Procedure

Dissolve 0.01mole(0.91gm) of thio semicarbazide in 10ml of warm water added 0.01 mole(1.40gm) of 3-Chlorobenzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated 1-(3-Chlorobenzylidene) thiosemicarbazide was filtered and dried. To 0.005mole(1.56gm) of 1(3Chlorobenzylidene)thiosemicarbazide in 300ml warm water, 0.01mole (1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with four portions of ammonia. The precipitated(5-(3-Chlorophenyl)- 1,3,4-thiadiazol-2-amine) was seperated by filteration and recrystallised from ethanol.

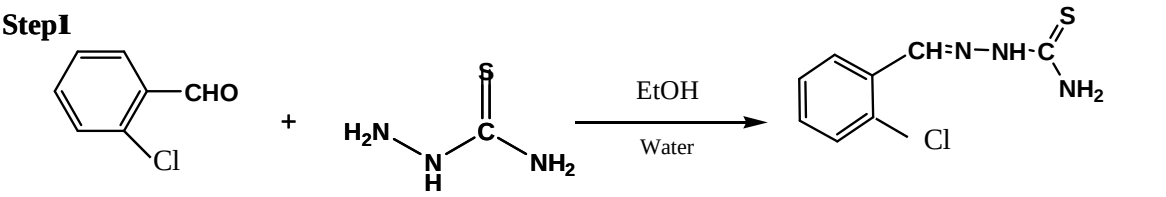
To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (2.75gm) of p-nitroaniline drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the coldwater. The precipitate 0.025mole(8.45gm) of N-[5-(3-Chlorophenyl)-1,3,4-thiadiazol-2yl] N-(4-Nitrophenyl)-methanediamine was filtered and dried. The product obtained was recrystallised from ethanol.

Synthesis of compound8B$_1$:

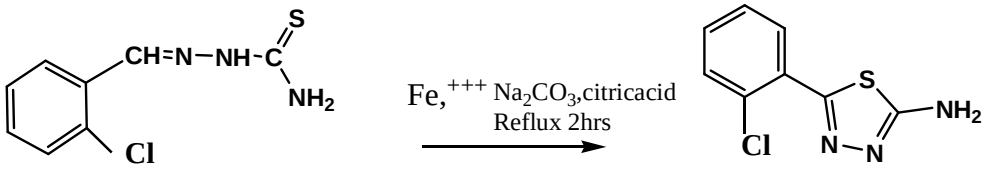
4-[({[5-(2-Chlorophenyl)-1,3,4-thiadiazolyl]amino}methyl)amino]benzenesulphonamide

StepI



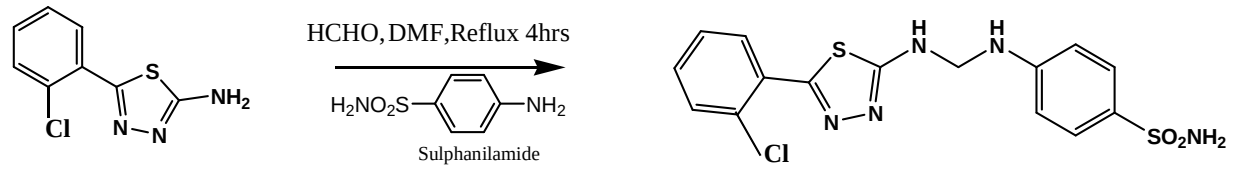
2-Chlorobenzaldehyde Thiosemicarbazide 1-(2-chlorobenzylidene)thiosemicarbazide

StepII



1-(2-chlorobenzylidene)thiosemicarbazide 5-(2-chlorophenyl)-1,3,4-thiadiazol-2-amine

StepIII



5-(2-chlorophenyl)-1,3,4-thiadiazol-2-amine 4-[({[5-(2-chlorophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulfonami

Procedure

Dissolve 0.01mole(0.91gm) of thio semicarbazide in 10ml of warm water added 0.01 mole(1.40gm) of 2-Chlorobenzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated 1-(2-Chlorobenzylidene)thiosemicarbazide was filtered and dried To 0.005 mole(0.106gm) of 1-(2-Chlorobenzylidene)thiosemicarbazide in 300ml warm water, 0.01mole (1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with four portions of ammonia. The precipitate(5-(2-Chlorophenyl)- 1,3,4-thiadiazol-2-amine) was seperated by filteration and recrystallised from ethanol.

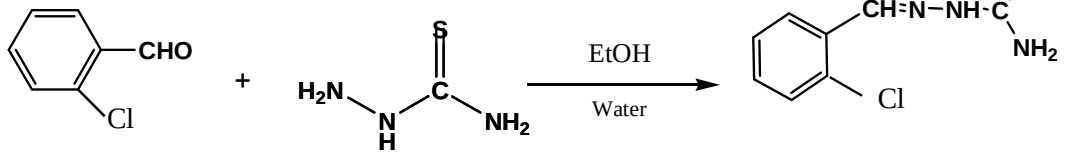
To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (3.45gm) of p-sulphanilamide drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the coldwater. The precipitate 0.025mole (8.95gm) of 4-[({[5-(2-Chlorophenyl)-1,3,4-thiadiazol-2yl]amino}methyl}amino benzene sulphonamide was filtered and dried. The product obtained was recrystallised from ethanol.

Synthesis of compound8B$_2$:

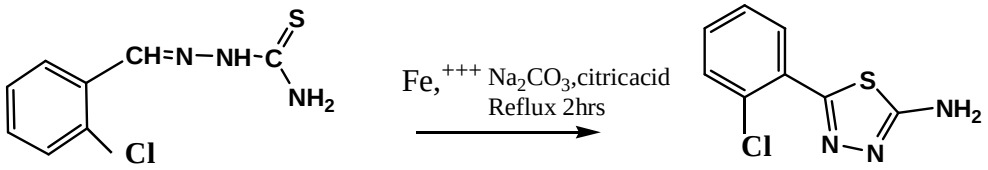
N-(4-Chlorophenyl)-N^1-[5-(2-Chlorophenyl)-1,3,4-thiadiazol-2-yl]methanediamine

StepI



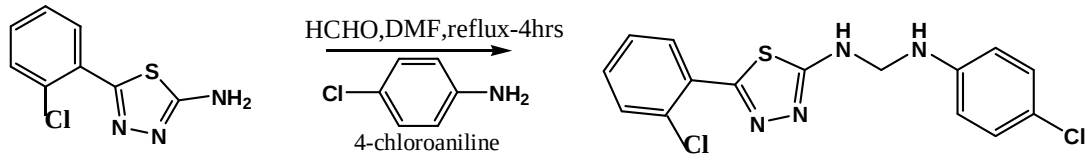
2-Chlorobenzaldehyde Thiosemicarbazide 1-(2-chlorobenzylidene)thiosemicarbazide

StepII



1-(2-chlorobenzylidene)thiosemicarbazide 5-(2-chlorophenyl)-1,3,4-thiadiazol-2-amine

StepIII



5-(2-chlorophenyl)-1,3,4-thiadiazol-2-amine *N*-(4-chlorophenyl)-*N'*-[5-(2-chlorophenyl)-1,3,4-thiadiazol-2-yl]methanediamine

Procedure

Dissolve 0.01mole(0.91gm) of thio semicarbazide in 10ml of warm water and 0.01 mole(1.40gm) of 2-Chlorobenzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated to form 1-(2-Chlorobenzylidene) thiosemicarbazid was filtered and dried .To

0.005mole(0.106gm) of 1 -(2-Chlorobenzylidene)thiosemicarbazide in 300ml warm water, 0.01mole (1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole (19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with four portions of ammonia. The precipitate(5-(2-Chlorophenyl)-1,3,4-thiadiazol-2-amine) was seperated by filteration and recrystallised from ethanol.

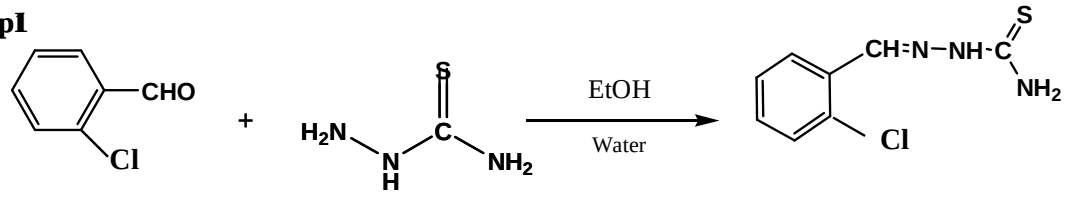
To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (2.55gm) of p-Chloroaniline drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the coldwater. The precipitate 0.025mole (8.95gm) of N-[5-(2-Chlorophenyl)-1,3,4-thiadiazol-2yl] N-(4-Chlorophenyl)-methanediamine was filtered and dried. The product obtained was recrystallised from ethanol.

Synthesis of compound $8B_3$:

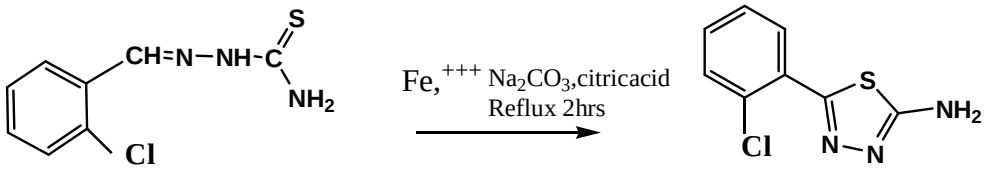
N-[5-(2-Chlorophenyl)-1,3,4-thiadiazol-2-yl]-N^1-(4-nitrophenyl)methanediamine

StepI



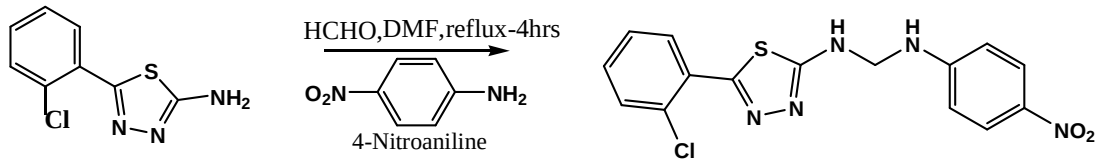
(2-Chlorobenzaldehyde) (Thiosemicarbazide) 1-(2-chlorobenzylidene)thiosemicarbazide

StepII



1-(2-chlorobenzylidene)thiosemicarbazide 5-(2-chlorophenyl)-1,3,4-thiadiazol-2-amine

StepIII



5-(2-chlorophenyl)-1,3,4-thiadiazol-2-amine

N-[5-(2-chlorophenyl)-1,3,4-thiadiazol-2-yl]- *N*'-(4-nitrophenyl)methanediamine

Procedure

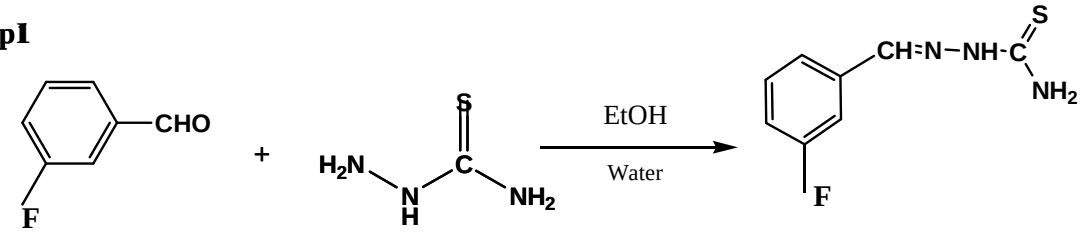
Dissolve 0.01mole (0.91gm)of thio semicarbazide in 10ml of warm water added 0.01 mole(1.40gm) of 2-Chlorobenzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated 1-(2-Chlorobenzylidene)thiosemicarbazide was filtered and dried. To 0.005 mole(0.106gm) of 1-(2-Chlorobenzylidene)thiosemicarbazide in 300ml warm water, 0.01mole (1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with four portions of ammonia ammonia. The precipitate(5-(2-Chlorophenyl)- 1,3,4-thiadiazol-2-amine) was seperated by filteration and recrystallised from ethanol.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (2.76gm) of p-nitroaniline drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the coldwater. The precipitate 0.025mole of (9.25gm) of N-[5-(2-Chlorophenyl)-1,3,4-thiadiazol-2yl] N-(4-Nitrophenyl)-methanediamine was filtered and dried. The product obtained was recrystallised from ethanol.

Synthesis of compound 8C_1:4-[({[5-(3-fluorophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulphonamide

Step1

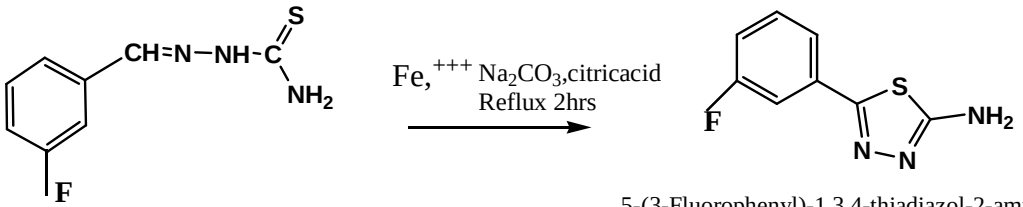


3-Fluorobenzaldehyde

Thiosemicarbazide

1-(3-Fluorobenzylidene)thiosemicarbazide

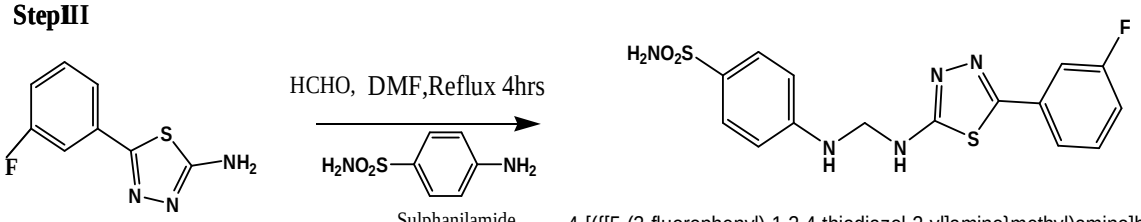
StepII



1-(3-Fluorobenzylidene)thiosemicarbazide

5-(3-Fluorophenyl)-1,3,4-thiadiazol-2-amine

StepIII



5-(3-Fluorophenyl)-1,3,4-thiadiazol-2-amine

Sulphanilamide

4-[({[5-(3-fluorophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulfonami

Procedure

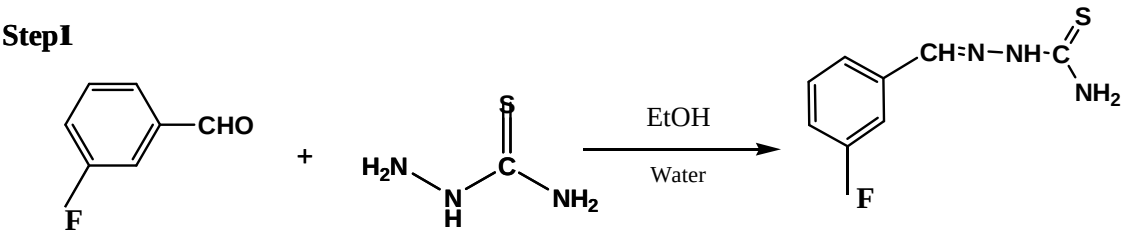
Dissolve 0.01mole (0.91gm)of thio semicarbazide in 10ml of warm water added 0.01 mole(1.24gm) of 3-fluorobenzaldehyde in 10ml of ethanol and stirred well for 10mts, the precipitated 1-(3-Fluorobenzylidene) thiosemicarbazide was filtered and dried. To 0.005 mole(0.98gm) of 1-(3-Fluorobenzylidene)thiosemicarbazide in 300ml warm water, 0.01mole (1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with . four portions of ammonia. The precipitate (5-(3-Fluorophenyl)- 1,3,4-thiadiazol-2-amine) was seperated by filteration and recrystallised from ethanol.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (3.45gm) of p-sulphanilamide drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and pouredintothecoldwater.Theprecipitate0.025mole(9.55gm)of4[({[5(3fluorophenyl)1,3,4thiadiazol2yl]amino}methyl)amino]benzenesulphonamidewascollected. was filtered and dried. The product obtained was recrystallised from ethanol.

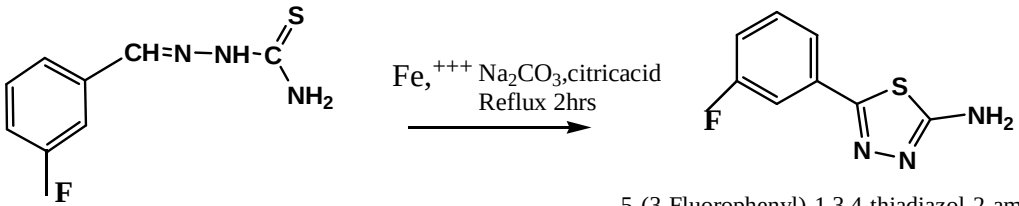
Synthesis of compound $8C_2$: N-(4-Chlorophenyl)-N^1-[5-3-fluorophenyl)-1,3,4-thiadiazol-2-yl]methanediamine

StepI



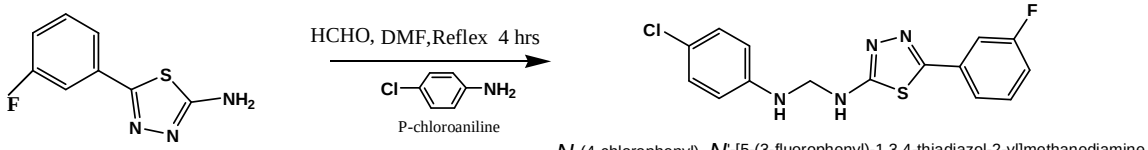
3-Fluorobenzaldehyde Thiosemicarbazide 1-(3-Fluorobenzylidene)thiosemicarbazide

StepII



1-(3-Fluorobenzylidene)thiosemicarbazide 5-(3-Fluorophenyl)-1,3,4-thiadiazol-2-amine

StepIII



5-(3-Flulorophenyl)-1,3,4-thiadiazol-2-amine *N*-(4-chlorophenyl)- *N'*-[5-(3-fluorophenyl)-1,3,4-thiadiazol-2-yl]methanediamine

Procedure

Dissolve 0.01mole(0.91gm) of thio semicarbazide in 10ml of warm water and 0.01 mole91.24gm) of 3-fluorobenzaldehyde in 10ml of ethanol to form 1-(3-Fluorobenzylidene) thiosemicarbazide. To 0.005 mole(0.98gm) of 1-(3-Fluorobenzylidene)thiosemicarbazide in 300ml warm water, 0.01mole (1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with ammonia. The precipitate(5-(3-Fluorophenyl)- 1,3,4-thiadiazol-2-amine) was seperated by filteration. ethanol and stirred well for 10mts, the precipitated was filtered and dried. four portions of ammonia. and recrystallised from ethanol.

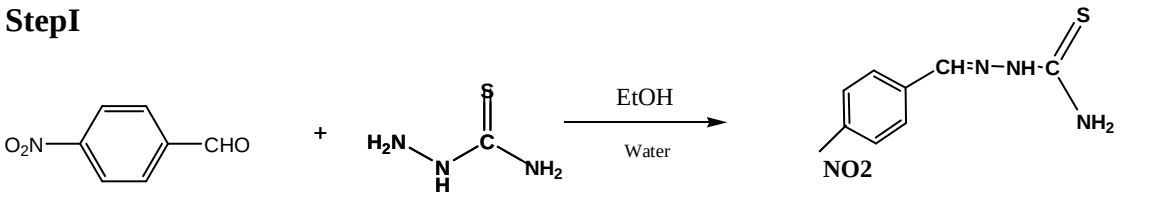
To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (2.55gm) of p-Chloroaniline drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the coldwater. The precipitate 0.025mole (8.95gm) of N-(4-Chlorophenyl)-N1-[5-(3-fluorophenyl)-1,3,4-thiadiazol-2-yl]methanediamine was filtered and dried. The product obtained was recrystallised from ethanol.

Synthesis of compound $8D_1$:

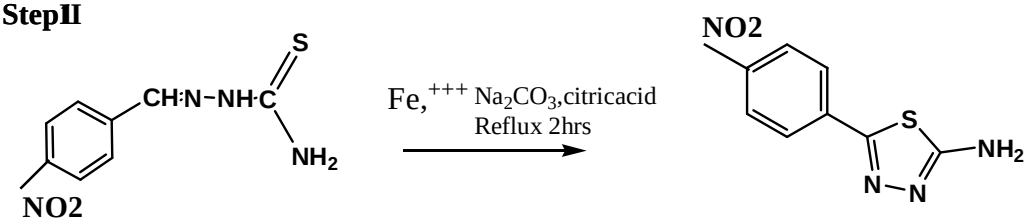
4-[({[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulphonamide

StepI



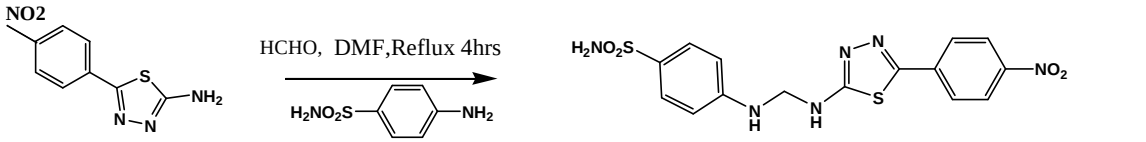
P-nitrobenzaldehyde Thiosemicarbazide 1-(4-nitrobenzylidene)thiosemicarbazide)

StepII



1-(4-nitrobenzylidene)thiosemicarbazide) 5-(4-nitrophenyl)-1,3,4-thiadiazol-2-amine

StepIII



5-(4-nitrophenyl)-1,3,4-thiadiazol-2-amine Sulphanilamide 4-[({[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulfonamide

Procedure

Dissolve 0.01mole(0.91gm) of thio semicarbazide in 10ml of warm water and 0.01 mole(1.510gm) of 4-nitrobenzaldehyde in 10ml of ethanol to form 1-(4-

nitrobenzylidene) thiosemicarbazide. To 0.005 mole (0.112gm) of 1-(4-nitrobenzylidene)thiosemicarbazide in 300ml warm water, 0.01mole(1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with ammonia. The precipitate(5-(4-nitrophenyl)- 1,3,4-thiadiazol-2-amine) was seperated by filteration.

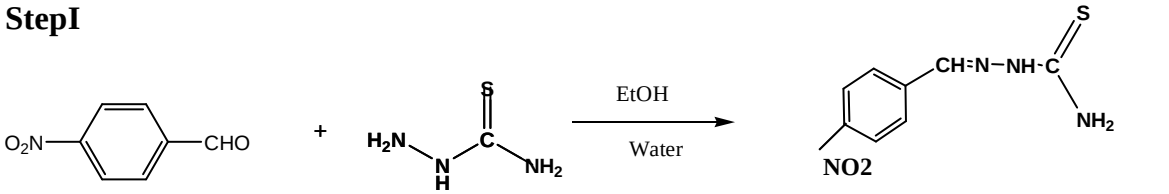
To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (3.45gm) of p-sulphanilamide drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the coldwater. The precipitate 0.025 mole(10.5gm) of 4-[({[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl}amino]benzenesulphonamide was filtered and dried. The product obtained was recrystallised from ethanol.

Synthesis of compound $8D_2$:

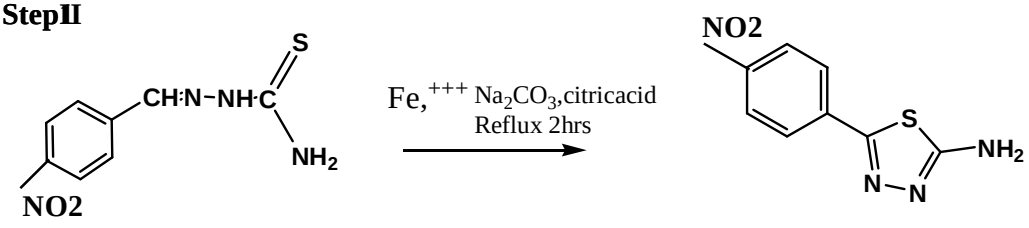
N-(4-nitrophenyl)-N-[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]methanediamine

StepI



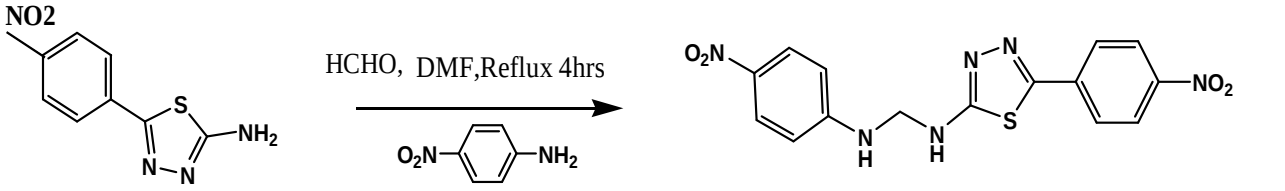
P-nitrobenzaldehyde Thiosemicarbazide 1-(4-nitrobenzylidene)thiosemicarbazide)

StepII



1-(4-nitrobenzylidene)thiosemicarbazide) 5-(4-nitrophenyl)-1,3,4-thiadiazol-2-amine

StepIII



5-(4-nitrophenyl)-1,3,4-thiadiazol-2-amine

p-nitroaniline

N-(4-nitrophenyl)-*N*'-[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]methanediamin

Procedure

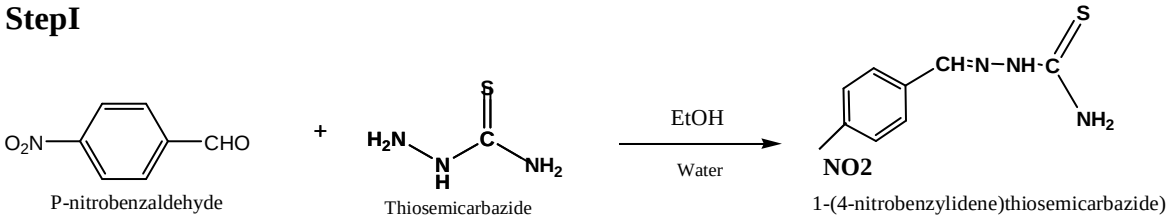
Dissolve 0.01mole (0.91gm) of thio semicarbazide in 10ml of warm water and 0.01 mole(1.51gm) of 4-nitrobenzaldehyde in 10ml of ethanol to form 1-(4-nitrobenzylidene)thiosemicarbazide.To 0.05mole(0.112gm)of 1(4nitrobenzylidene)thio semicarbazide in 300ml warm water, 0.01mole(1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole(5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with ammonia. The precipitate(5-(4-nitrophenyl)- 1,3,4-thiadiazol-2-amine) was seperated by filteration.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (2.76gm) of p-nitroaniline drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the coldwater. The precipitate 0.025mole (9.754gm) of N-(4-nitrophenyl)-N-[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]methanediamine was filtered and dried. The product obtained was recrystallised from ethanol.

Synthesis of compound $8D_3$: N-(4-Chlorophenyl)-N^1-[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]methanediamine

StepI

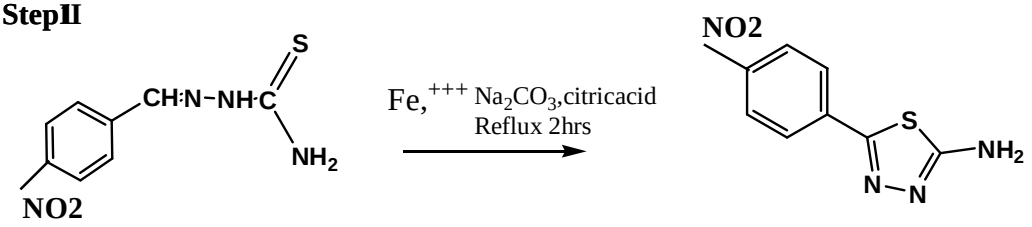


P-nitrobenzaldehyde

Thiosemicarbazide

1-(4-nitrobenzylidene)thiosemicarbazide)

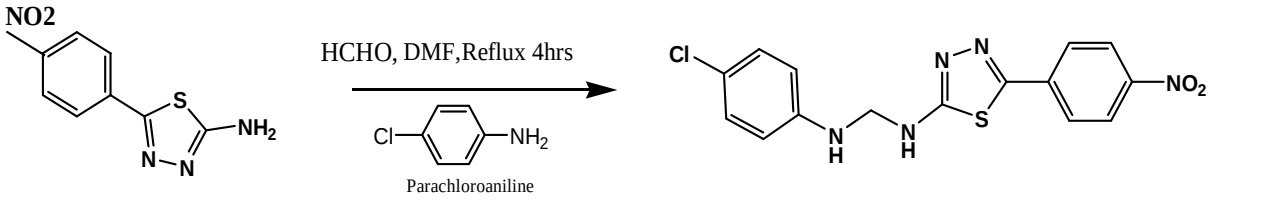
StepII



1-(4-nitrobenzylidene)thiosemicarbazide)

5-(4-nitrophenyl)-1,3,4-thiadiazol-2-amine

StepIII



5-(4-nitrophenyl)-1,3,4-thiadiazol-2-amine

N-(4-chlorophenyl)-*N'*-[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]methanediamin

Procedure

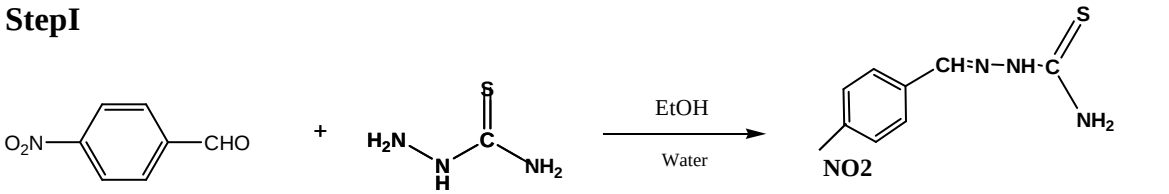
Dissolve 0.01mole(0.91gm) of thio semicarbazide in 10ml of warm water and 0.01 mole(1.510gm) of 4-nitrobenzaldehyde in 10ml of ethanol to form 1-(4-nitrobenzylidene) thiosemicarbazide. To 0.005 mole(0.112gm) of 1-(4-nitrobenzylidene)thiosemicarbazide in 300ml warm water, 0.01mole(1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with ammonia. The precipitate(5-(4-nitrophenyl)- 1,3,4-thiadiazol-2-amine) was seperated by filteration.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (2.76gm) of p-chloroaniline drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the coldwater. The precipitate 0.025mole (9.75gm) of N-(4-chlorophenyl)-N-[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]methanediamine was filtered and dried. The product obtained was recrystallised from ethanol.

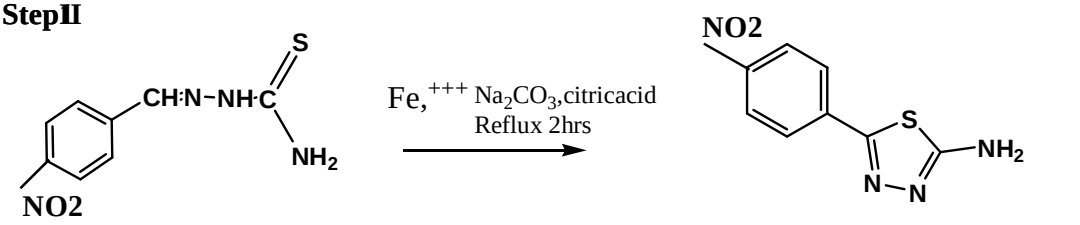
Synthesis of compound 8D$_4$: N-(4-methoxyphenyl)-N^1-[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]methanediamine

StepI



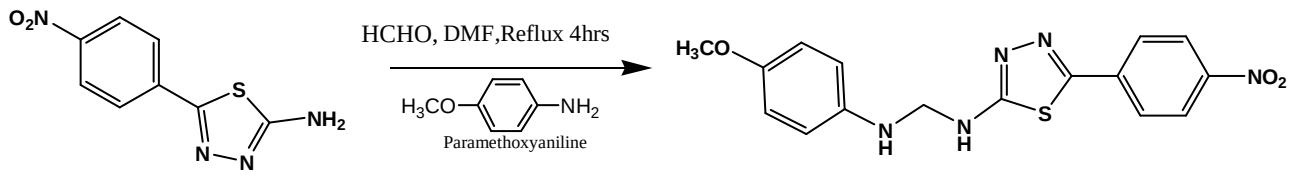
P-nitrobenzaldehyde Thiosemicarbazide 1-(4-nitrobenzylidene)thiosemicarbazide)

StepII



1-(4-nitrobenzylidene)thiosemicarbazide) 5-(4-nitrophenyl)-1,3,4-thiadiazol-2-amine

StepIII



5-(4-nitrophenyl)-1,3,4-thiadiazol-2-amine *N*-(4-methoxyphenyl)-*N*'-[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]methanediamir

Procedure

Dissolve 0.01mole(0.91gm) of thio semicarbazide in 10ml of warm water and 0.01 mole(1.510gm) of 4-nitrobenzaldehyde in 10ml of ethanol to form 1-(4-nitrobenzylidene) thiosemicarbazide. To 0.005 mole(0.112gm) of 1-(4-nitrobenzylidene)thiosemicarbazide in 300ml warm water, 0.01mole (1.62gm) of Ferric chloride in 300ml water was added slowly with stirring. The contents were refluxed for 2hrs at 100^0C. The solution was filtered hot and added 0.1 mole(19.2gm) of citricacid with 0.05mole (5.25gm) of sodiumcarbonate. The resulting mixture was neutralised with ammonia. The precipitate(5-(4-nitrophenyl)- 1,3,4-thiadiazol-2-amine) was seperated by filteration.

To the above synthesized compound(0.03mole) added 15ml of DMF followed by added 1ml of formaldehyde(37-48%) and 0.02 mole (2.56gm) of p-methoxyaniline drop by drop with stirring.

The reaction mixture was stirred well and refluxed for 4-5hrs and poured in to the coldwater. The precipitate 0.025mole(9.25gm) of N-(4-methoxyphenyl)-N-[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]methanediamine was filtered and dried. The product obtained was recrystallised from ethanol.

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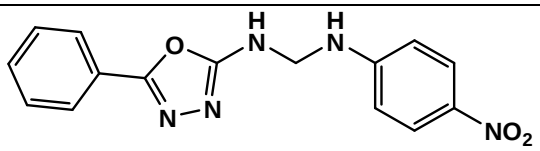
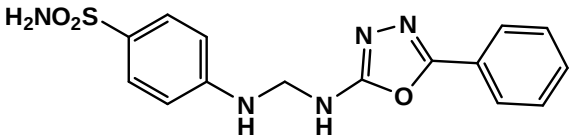
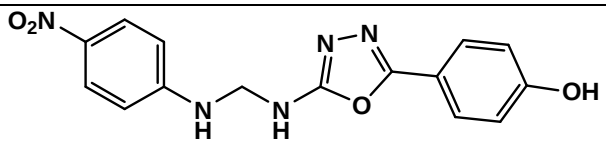
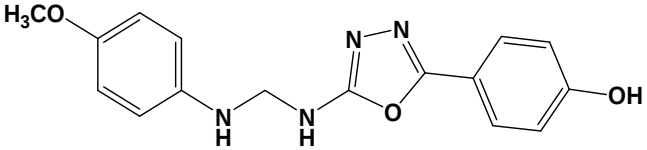
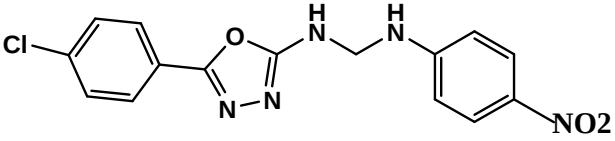
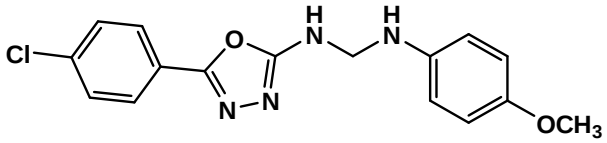
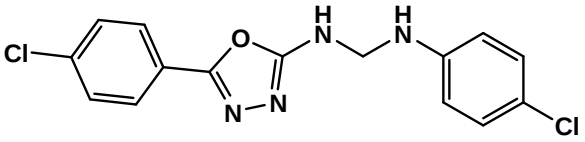
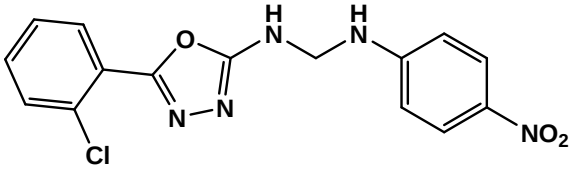
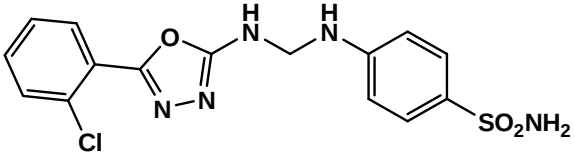
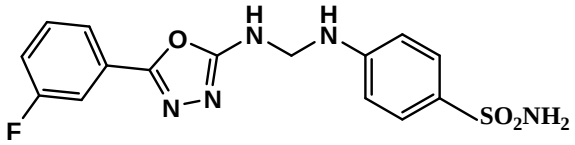
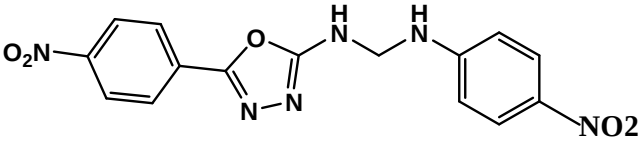
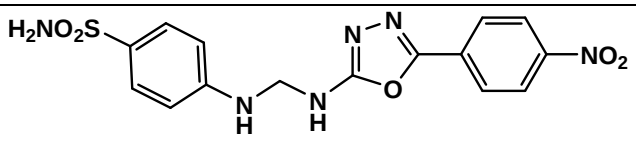
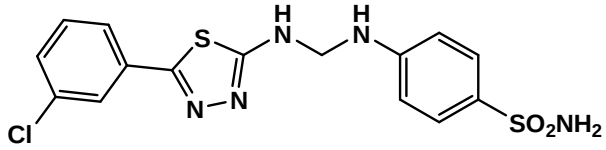
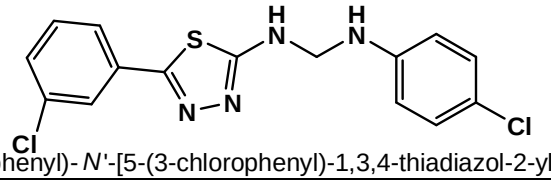
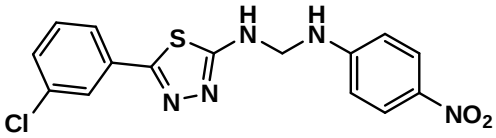
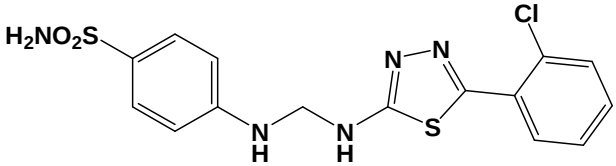
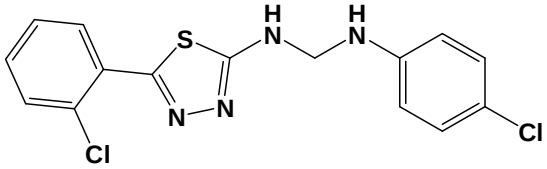
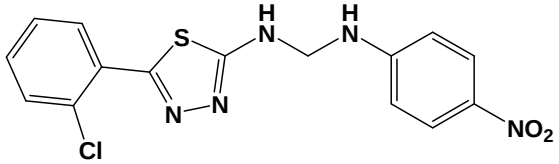
LIST OF COMPOUNDS SYNTHESIZED

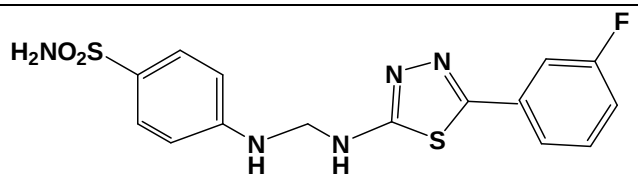
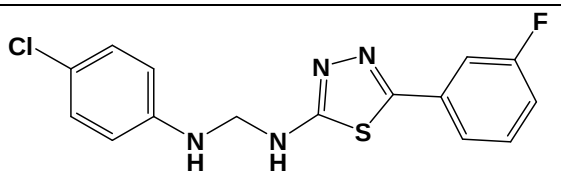
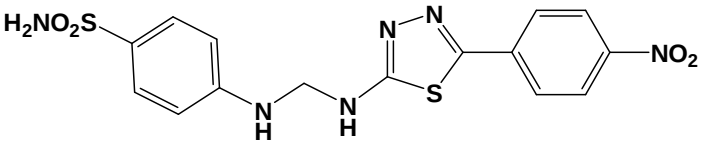
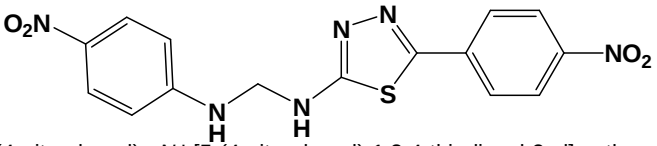
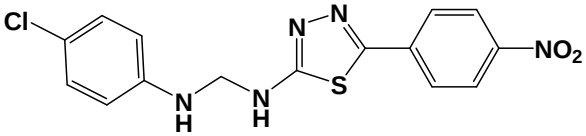
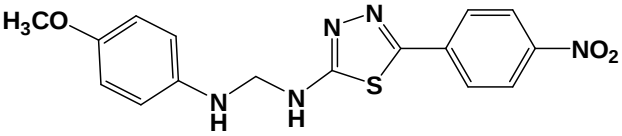
List of compounds synthesized

Table no:1

Serial no	Compound code	Structure of compounds and chemical name
1	1b	 *N*-((4-nitrophenylamino)methyl)-5-phenyl-1,3,4-oxadiazol-2-amine
2	1e	 4-({[(5-phenyl-1,3,4-oxadiazol-2-yl)amino]methyl}amino)benzenesulfonamide
3	2b	 4-(5-((4-nitrophenylamino)methylamino)-1,3,4-oxadiazol-2-yl)phenol
4	2c	 4-[5-({[(4-methoxyphenyl)amino]methyl}amino)-1,3,4-oxadiazol-2-yl]phenol
5	3b	 *N* -[5-(4-chlorophenyl)-1,3,4-oxadiazol-2-yl]- *N*'-(4-methylphenyl)methanediamine

6	**3c**	 *N*-((4-methoxyphenylamino)methyl)-5-(4-chlorophenyl)-1,3,4-oxadiazol-2-amine
7	**3d**	 *N* -(4-chlorophenyl)- *N*'-[5-(4-chlorophenyl)-1,3,4-oxadiazol-2-yl]methanediamine
8	**4b**	 *N*-[5-(2-chlorophenyl)-1,3,4-oxadiazol-2-yl]- *N*'-(4-nitrophenyl)methanediamine
9	**4e**	 4-[({[5-(2-chlorophenyl)-1,3,4-oxadiazol-2yl]amino}methyl)amino]benzenesulfonamide
10	**5e**	 N-(4-Sulphonamyl)-N-[5-(3-Fluorophenyl)-1,3,4-oxadiazol-2-yl]methanediamine
11	**6b**	 N-[5-(4-nitrophenyl)-1,3,4-oxadiazol-2-yl]-N1-nitrophenylmethanediamine

12	**6e**	 4-[({[5-(4-nitrophenyl)-1,3,4-oxadiazol-2-yl]amino}methyl)amino]benzenesulfonamide
13	**8a1**	 4-[({[5-(3-chlorophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulfonamide
14	**8a2**	 *N* -(4-chlorophenyl)-*N*'-[5-(3-chlorophenyl)-1,3,4-thiadiazol-2-yl]methanediamine
15	**8a3**	 *N*-[5-(3-chlorophenyl)-1,3,4-thiadiazol-2-yl]-*N*'-(4-nitrophenyl)methanediamine
16	**8b1**	 4-[({[5-(2-chlorophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulfonamide
17	**8b2**	 *N* -(4-chlorophenyl)- *N*'-[5-(2-chlorophenyl)-1,3,4-thiadiazol-2-yl]methanediamine
18	**8b3**	 *N*-[5-(2-chlorophenyl)-1,3,4-thiadiazol-2-yl]-*N*'-(4-nitrophenyl)methanediamine

19	**8c1**	 4-[({[5-(3-fluorophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulfonamide
20	**8c2**	 *N* -(4-chlorophenyl)- *N* '-[5-(3-fluorophenyl)-1,3,4-thiadiazol-2-yl]methanediamine
21	**8d1**	 4-[({[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulfonamide
22	**8d2**	 *N* -(4-nitrophenyl)- *N* '-[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]methanediamine
23	**8d3**	 *N* -(4-chlorophenyl)- *N* '-[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]methanediamine
24	**8d4**	 *N* -(4-methoxyphenyl)*N* '-[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]methanediamine

IDENTIFICATION AND CHARACTERIZATION

Identification and characterization

Solubility

Table no:2

Compound code.	Soluble in	Insoluble in
1b	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
1e	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
2b	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
2c	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
3b	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
3c	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
3d	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
4b	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
4e	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
5e	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
6b	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
6e	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
$8a_1$	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
$8a_2$	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
$8a_3$	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
$8b_1$	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
$8b_2$	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
$8b_3$	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
$8c_1$	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
$8c_2$	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
$8d_1$	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
$8d_2$	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
$8d_3$	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water
$8d_4$	DMSO, DMF, ethanol, chloroform, acetone	CCl_4,water

Thin layer chromatography

Chromatography is an important technique to identify the formation of new compounds and also to determine the purity of the compound. The Rf value is characteristic for each of the compound.

a. **Preparation of chromatoplate:**

   Cleaned and dried glass plates were taken. Uniform slurry of silica Gel-G in water was prepared in the ratio 1:2. The slurry was then poured into the chamber of TLC applicator,

   Glass plate were moved under the applicator smoothly to get an uniform coating of slurry on the plates.

   The plates were dried first at room temperature and then kept for activation at 110^0C for 1hour.

b. **Preparation of solvent system and saturation of chamber:**

   The solvent system used for development of chromatogram was prepared carefully by mixing.

   Methanol: chloroform [1:1]

c. **Application of sample:**

   The solution of parent compounds and its target molecule were taken in small bored capillary tube and spotted at 2centimteers from the base end of the plate. After spotting the plate will allow to dry at room temperature and plates were transferred to chromatogrophic chamber containing solvent system for development.

d. **Development of chromatogram:**

   Plates were developed by ascendind technique when solvent front had reached a disance of 10-12centimeters; they were taken out and dried at room temperature.

e. **Detection of spots.**

   The developed spots were detected by exposing them to iodine vapours.

f. **Calculation of Rf value:**

   Formula for calculating Rf value of synthesized compounds.

   Rf value = Distance travelled by sample / Distance travelled by solvent front.

Table no:3

Compound code	Mobile Phase	Ratio	Rf value
1b	Methanol: Chloroform	1:1	0.54
1e	Methanol: Chloroform	1:1	0.77
2b	Methanol: Chloroform	1:1	0.56
2c	Methanol: Chloroform	1:1	0.70
3b	Methanol: Chloroform	1:1	0.59
3c	Methanol: Chloroform	1:1	0.66
3d	Methanol: Chloroform	1:1	0.60
4b	Methanol: Chloroform	1:1	0.69
4e	Methanol: Chloroform	1:1	0.66
5e	Methanol: Chloroform	1:1	0.5
6b	Methanol: Chloroform	1:1	0.87
6e	Methanol: Chloroform	1:1	0.64
$8a_1$	Methanol: Chloroform	1:1	0.73
$8a_2$	Methanol: Chloroform	1:1	0.53
$8a_3$	Methanol: Chloroform	1:1	0.73
$8b_1$	Methanol: Chloroform	1:1	0.65
$8b_2$	Methanol: Chloroform	1:1	0.65
$8b_3$	Methanol: Chloroform	1:1	0.74
$8c_1$	Methanol: Chloroform	1:1	0.67
$8c_2$	Methanol: Chloroform	1:1	0.56
$8d_1$	Methanol: Chloroform	1:1	0.54
$8d_2$	Methanol: Chloroform	1:1	0.76
$8d_3$	Methanol: Chloroform	1:1	0.65
8d4	Methanol: Chloroform	1:1	0.76

Physical properties of the synthesized compounds

Table no:4

Compound Code	Melting.pt.(°C)	% Yield	Molecular formula	Molecular Weight(gm)
1b	230-35	90%	$C_{15}H_{13}N_5O_3$	311.31
1e	225-30	89%	$C_{15}H_{15}N_5O_3S$	345.38
2b	230-35	90%	$C_{15}H_{13}N_5O_4$	327.29
2c	240-45	88%	$C_{16}H_{16}N4O_3$	312.32
3b	235-40	91%	$C_{15}H_{12}ClN_5O_3$	381.45
3c	230-35	86%	$C_{16}H_{15}ClN_4O_2$	335.12
3d	210-15	89%	$C_{15}H_{12}C_{l2}N_4O$	312.13
4b	215-20	91%	$C_{15}H_{12}ClN_4O_2$	345.73
4e	240-45	90%	$C_{15}H_{14}ClN_5O_3S$	372.83
5e	230-35	83%	$C_{15}H_{14}FN_5O_3S$	363.37
6b	230-35	90%	$C_{16}H_{15}N_6O_5$	371.33
6e	215-20	86%	$C_{15}H_{14}N6O_5S$	367.60
$8a_1$	220-25	89%	$C_{15}H_{14}ClN_5O_3S$	379.82
$8a_2$	210-15	90%	$C_{15}H_{12}C_{l2}N_4O$	335.19
$8a_3$	225-30	91%	$C_{15}H_{12}ClN_5O_2S$	361.81
$8b_1$	230-35	92%	$C_{15}H_{14}ClN_5O_2S_2$	395.89
$8b_2$	225-30	89%	$C_{15}H_{12}C_{l2}N_4S$	351.25
$8b_3$	220-25	90%	$C_{15}H_{12}ClN_5O_2S$	361.81

$8c_1$	230-35	86%	$C_{15}H_{14}ClN_5O_2S_2$	379.18
$8c_2$	220-25	88%	$C_{15}H_{12}C_{l2}N_4S$	351.25
$8d_1$	215-20	89%	$C_{15}H_{14}N_6O_4S_2$	406.44
$8d_2$	220-25	88%	$C_{15}H_{12}N6O_4S$	372.36
$8d_3$	210-15	90%	$C_{15}H_{12}ClN_5O_2S$	361.81
$8d_4$	220-25	85%	$C_{16}H_{15}N_5O_3S$	357.39

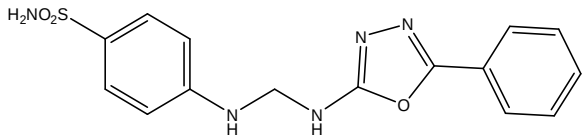
SPECTRAL STUDIES

IR Spectra

The peak in IR spectrum gives an idea about the functional group and probable structure of the compound in IR region ranges between 4000-666 $cm^{-1.}$ Quanta of radiation from this region of the spectrum correspond to energy differences between different vibrational levels of molecules.The compound was recorded on Jasco FTIR-4100 spectrometer shows different vibration levels of molecules by using KBr disk technique. The IR spectrum of compound as follows:

Characteristics IR absorption of compounds

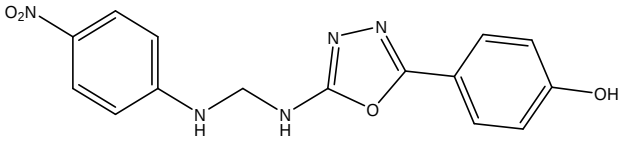
Compound code: 1e



4-({[(5-phenyl-1,3,4-oxadiazol-2-yl)amino]methyl}amino)benzenesulfonamide

N-H stretching =3413cm-1, Aromatic Stretching = 2849,1574,750cm-1,
C-N stretching = 1470cm-1, C- O-C stretching = 1094cm-1

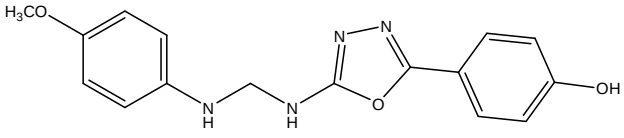
Compound code: 2b



4-(5-((4-nitrophenylamino)methylamino)-1,3,4-oxadiazol-2-yl)phenol

N-H stretching vibration = 3374 cm-1, Aromatic stretching=2918,1574,738 cm-1
,N=O Stretching=1512,1344 cm-1 C-O-C stretching = 1043cm-1

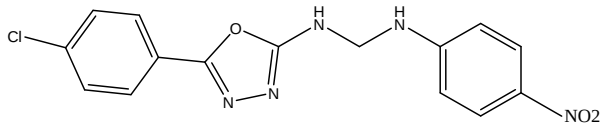
Compound code: 2c



4-[5-({[(4-methoxyphenyl)amino]methyl}amino)-1,3,4-oxadiazol-2-yl]phenol

OH Stretching vibration=3264 cm-1,N-H Stretching =3173 cm-1,Aromatic stretching=2873,1654,822cm-1,C-N Stretching=1387 cm-1,C-O-CStretching=1092cm-1

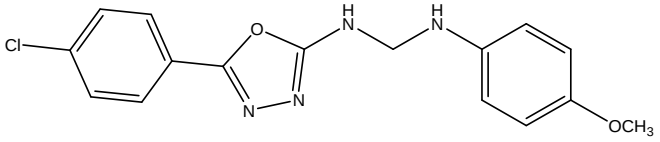
Compound code: 3b



N-[5-(4-chlorophenyl)-1,3,4-oxadiazol-2-yl]- *N*'-(4-methylphenyl)methanediamine

N-H Stretching=3353 cm-1,Aromatic stretching=2952 cm-1,1688cm-1,763 cm-1,
N=O Stretching=1566 cm-1,1372 cm-1,C-N Stretching=1435 cm-1,
C-O-C Stretching=1234 cm-1,C-Cl=962 cm-1

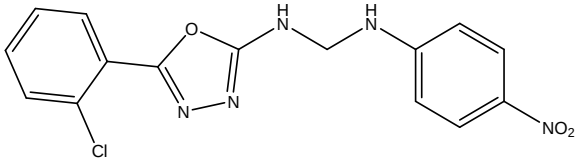
Compound code: 3c



N-((4-methoxyphenylamino)methyl)-5-(4-chlorophenyl)-1,3,4-oxadiazol-2-amine

N-H Stretching=3430 cm-1,Aromatic Stretching=3310 cm-1,1572 cm-1,740cm-1,
C-N Stretching=1423 cm-1,C-O-C Stretching=1161 cm-1,

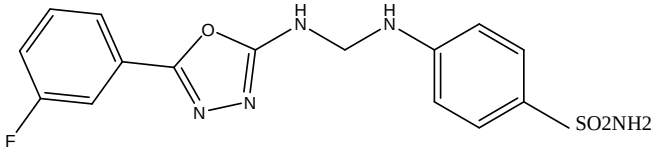
Compound code: 4b



N-[5-(2-chlorophenyl)-1,3,4-oxadiazol-2-yl]- *N*'-(4-nitrophenyl)methanediamine

N-H Stretching=3373 cm-1,Aromatic Stretching=3085 cm-1157 cm-1,738cm-1,
N=O stretching=1511 cm-1,C-O-C Stretching=1043cm-1,C-Cl=862cm-1

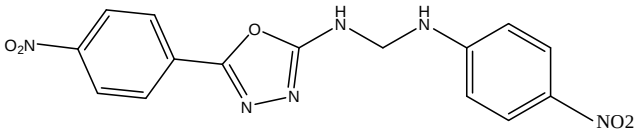
Compound code: 5e



N-(4-Sulphonamyl)-N-[5-(3-Fluorophenyl)-1,3,4-oxadiazol-2-yl]methanediamine

N-H Stretching=3100 cm-1,Aromatic Stretching=1648 cm-1,821 cm-1,
C-F=1387 cm-1,C-N=1319 cm-1,C-O-C Stretching=1092 cm-1,S=O=1050 cm-1

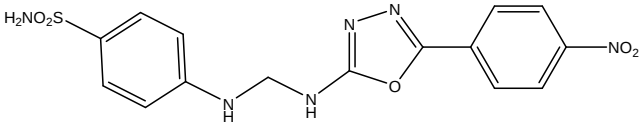
Compound code: 6b



N-[5-(4-nitrophenyl)-1,3,4-oxadiazol-2-yl]-N1-nitrophenylmethanediamine

N-H Stretching=3374cm-1,Aromatic Stretching=2918cm-1,1618cm-1,
738cm-1,N=OStretching=1575cm-1,1329cm-1,C-NStretching=1424cm-1,
C-O-C Stretching=1043cm-1

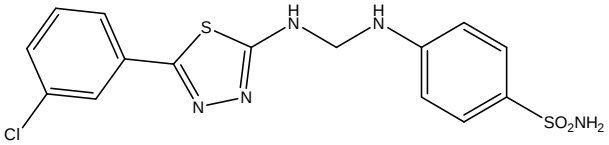
Compound code: 6e



4-[({[5-(4-nitrophenyl)-1,3,4-oxadiazol-2-yl]amino}methyl)amino]benzenesulfonamide

N-H Stretching=2926cm-1,Aromatic Stretching=1597 cm-1,752cm-1
,N=O Stretching=1507cm-1,1323cm-1,C-N Stretching=1459cm-1,S=O=1050cm-1
,C-O-C=1096cm-1

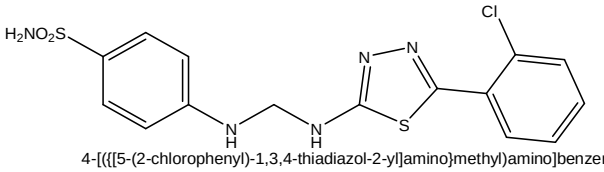
Compound code: $8a_1$



4-[({[5-(3-chlorophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulfonamide

Aromatic Stretching=1597cm-1,751cm-1.C-N Stretching=1374cm-1,S=O=1096cm-1,C-
Cl=815 cm-1,
C-S-C=676 cm-1

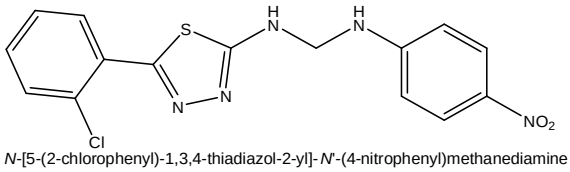
Compound code: $8b_1$



4-[({[5-(2-chlorophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulfonamide

N-H Sretching=3136cm-1,Aromatic Stretching=2926 cm-1,1596 cm-1,688cm-1,
C-N Stretching=1374cm-1,S=O=1050cm-1,C-Cl=822cm-1,C-S-C=688cm-1

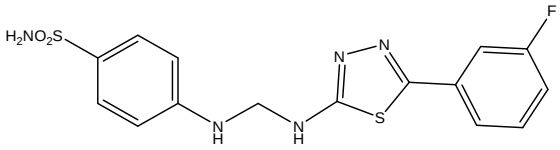
Compound code: $8b_3$



N-[5-(2-chlorophenyl)-1,3,4-thiadiazol-2-yl]-*N*'-(4-nitrophenyl)methanediamine

N-H Stretching=3373cm-1,Aromatic Stretchihg=1619cm-1,738cm-1, N=O=1574 cm-1329cm-1,C-N=1424cm-1,C-Cl=862cm-1,C-S-C=718cm-1

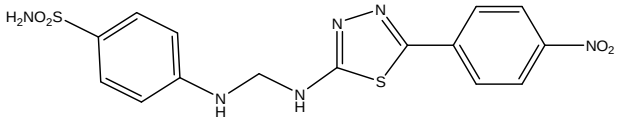
Compound code: $8c_1$



4-[({[5-(3-fluorophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulfonamide

N-H Stretching=3100cm-1,Aromatic Stretching=2920cm1,1595cm1,C-F=1450cm-1, C- N=1318cm-1,C=S=1091cm-1, C-S-C=908 cm-1

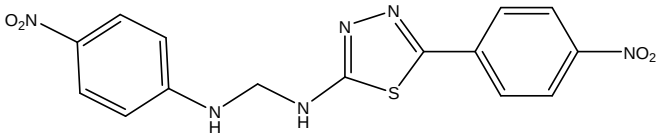
Compound code: $8d_1$



4-[({[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulfonamide

N-H Stretching=3100cm-1,Aromatic Stretching=2920 cm-1,1596 cm-1,815cm-1, N=O=1520 cm-1,1323 cm-1,S=O=1151 cm-1,C-S-C=750 cm-1

Compound code: $8d_2$



N-(4-nitrophenyl)-*N*'-[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]methanediamine

Aromatic nitro = 1542.77cm-1,N-H Stretching-3373cm-1, Aromatic Stretching=2920 cm-1,1618cm1,738cm-1, N=O=1575 cm-1,1329cm-1,C-N=1448cm-1,C-S-C=862cm-1

^{1}HNMR Spectr

NMR spectroscopy enables us to record differences in magnetic properties of the various magnetic nuclei present, and to deduce in large measure about the position of these nuclei are within the molecule. We can deduce how many different kinds of environment are there in the molecules and also which atom is present in neighboring groups. The proton NMR spectra enables us to know different chemical and Magnetic environment corresponding to proton in molecules.

Table no.6

Ser. No.	Comp. code	Hydrogen	δ(ppm)	Multiplicity	Solvent
1	**1b**	(2H,Ar-H,Ortho to nitro)	7.93-7.95	Doublet	DMSO
		(5H,Ar-H)	7.49-7.52	Multiplet	
		(2H,Ar-H)	6.90-6.91	Doublet	
		(1H,Ar-NH)	6.72	Singlet	
		(2H,CH_2)	3.53	Singlet	
		(1H,Oxazole-NH)	2.50	Singlet	
2	**4e**	(1H,Ar-NH)	7.95	Singlet	DMSO
		(4H,Ar-H)	7.80-7.81	Multiplet	
		(4H,Ar-H)	7.78-7.79	Multiplet	
		(4H,Ar-H)	7.78-7.79	Multiplet	
		(2H,NH_2)	7.65	Singlet	
		(1H,Oxazole-NH)	2.89	Singlet	
3	**6b**	(4H,Ar-H)	8.73-8.75	Doublet	DMSO
		(4H,Ar-H)	8.03-8.05	Doublet	
		(1H,Ar-NH)	6.73-6.76	Singlet	
		(2H,CH_2)	3.34	Singlet	
		(1H,Oxazole-NH)	2.50	Singlet	

4	**6e**	(4H,Ar-H)	7.40-7.61	Multiplet	DMSO
		(4H,Ar-H)	7.29	Multiplet	
		(1H,Ar-NH)	6.74	Singlet	
		(2H,CH_2)	3.34	Singlet	
		(1H,Oxazole-NH)	2.89	Singlet	
5	**8d$_1$**	(4H,Ar-H)	8.41-8.437	Doublet	DMSO
		(4H,Ar-H)	8.16-8.18	Doublet	
		(1H,Ar-NH)	7.958	Singlet	
		(2H,NH_2)	7.95	Singlet	
		(2H,CH_2)	3.34	Singlet	
		(1H,Oxazole-NH)	2.89	Singlet	

Mass spectroscopy

Mass spectroscopy enables us to know;

Relative molecular masses (molecular weight) with very high accuracy, from this exact molecular formula can be deduced. To detect within the molecule the places at which it prefers fragmentation, from these we can deduce the presence of recognizable groups within the molecule. As a method of identifying analytes by comparison of their mass spectra with libraries of digitalized mass spectra of known compound.

The spectral data of synthesized compounds as follows:

Table no.7

Sr. no.	Compound code	Molecular mass (in gram)
1	1b	311.03
2	4e	372.8
3	6b	396.2
4	6e	390.6
5	$8a_1$	379.82
6	$8b_3$	361.81
7	$8c_1$	379.18
8	$8d_1$	406.44

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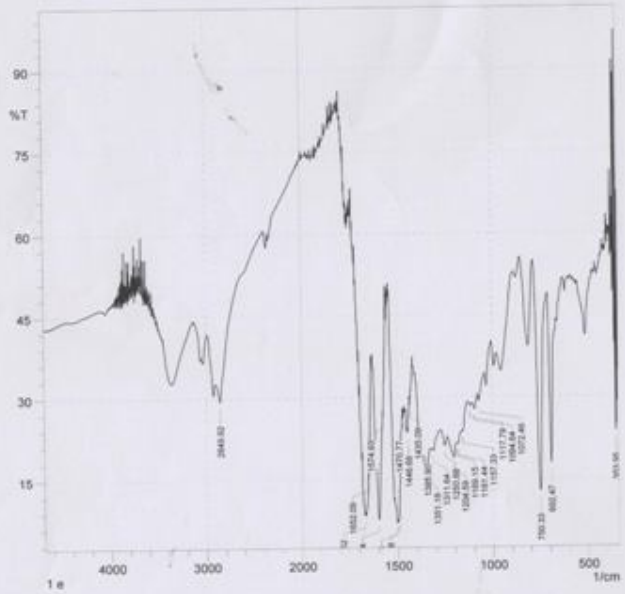
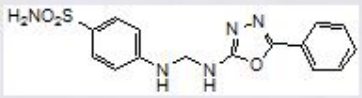


	Peak	Intensity	Corr. Inte	Base (H)	Base (L)	Area	Corr. Are
1	353.95	23.651	50.693	354.91	352.02	1.513	0.797
2	662.47	17.878	28.161	706.86	670.29	18.87	5.613
3	750.33	12.747	39.507	783.13	709.83	35.548	14.615
4	1072.46	29.195	1.963	1078.24	1041.8	18.235	0.331
5	1094.64	27.77	1.888	1108.14	1079.21	15.635	0.392
6	1117.79	28.35	0.647	1130.32	1109.11	11.498	0.096
7	1157.33	23.669	0.818	1161.19	1131.29	17.235	0.118
8	1181.44	21.49	0.394	1185.3	1162.15	14.877	0.046
9	1189.15	21.395	0.094	1193.01	1185.3	5.16	0.006
10	1204.59	18.894	2.899	1238.34	1193.98	30.444	1.356
11	1250.88	21.109	2.067	1272.1	1239.31	21.378	0.651
12	1311.64	20.161	0.338	1313.57	1275.95	24.367	0.027
13	1351.18	16.692	3.034	1363.72	1328.03	26.381	1.393
14	1385.9	21.872	3.483	1399.4	1380.11	11.658	0.592
15	1435.09	28.614	1.938	1437.02	1430.26	3.552	0.107
16	1446.66	23.523	1.503	1448.59	1437.98	6.227	0.138
17	1470.77	26.293	1.701	1472.7	1465.95	3.835	0.104
18	1494.88	7.662	0.937	1495.85	1476.56	15.25	0.035
19	1504.53	7.098	1.102	1507.42	1502.6	5.44	0.189
20	1574.93	26.427	2.453	1576.86	1570.11	3.664	0.155
21	1599.04	7.691	23.489	1624.12	1576.86	37.477	13.499
22	1652.09	15.125	2.098	1653.05	1636.65	10.201	0.174
23	1667.52	8.395	1.391	1669.45	1654.01	15.246	0.976
24	2849.92	29.381	6.863	2899.11	2618.45	107.859	4.092



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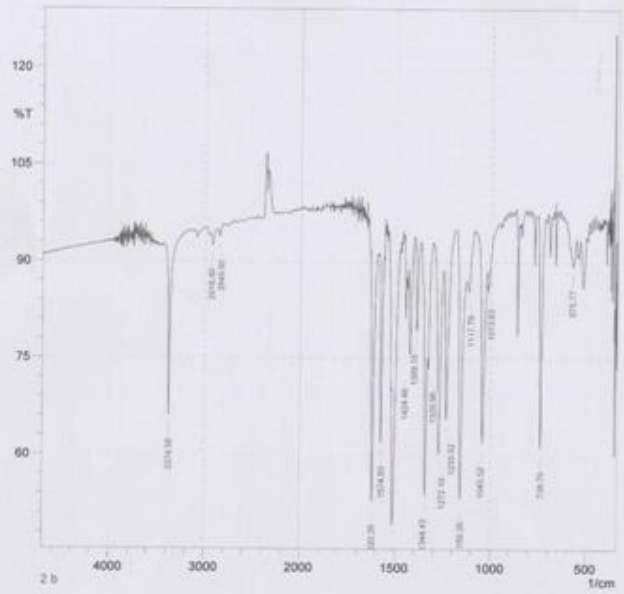
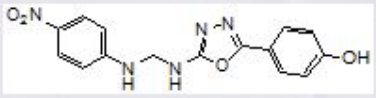


	Peak	Intensity	Corr. Inte	Base (H)	Base (L)	Area	Corr. Are
1	575.77	89.393	3.315	587.34	558.41	1.206	0.278
2	738.76	61.202	21.644	744.55	720.44	2.021	0.709
3	1013.63	86.432	0.268	1014.59	968.3	1.449	0.004
4	1043.52	61.905	28.89	1077.28	1024.24	4.679	2.639
5	1117.79	85.417	3.873	1127.43	1090.78	1.711	0.244
6	1159.26	53.374	38.176	1184.33	1128.39	6.132	3.845
7	1233.52	65.48	24.922	1250.88	1185.3	4.625	2.244
8	1272.1	60.208	30.104	1305.85	1251.84	5.338	3.073
9	1329.96	73.44	2.855	1331.89	1306.82	1.603	0.055
10	1344.43	53.831	27.161	1367.58	1332.86	4.727	1.969
11	1389.76	79.355	13.489	1398.44	1377.22	1.322	0.625
12	1424.48	74.011	16.584	1435.09	1404.22	2.168	0.971
13	1512.24	49.278	24.584	1519.96	1479.45	6.409	2.562
14	1574.93	61.896	29.25	1588.43	1562.39	2.637	1.589
15	1620.26	52.903	38.63	1634.73	1598.08	4.665	3.241
16	2849.92	93.811	1.26	2865.35	2817.13	1.109	0.087
17	2918.4	92.419	2.134	2942.51	2890.43	1.456	0.194
18	3374.58	66.002	26.697	3418.94	3296.46	7.453	3.446



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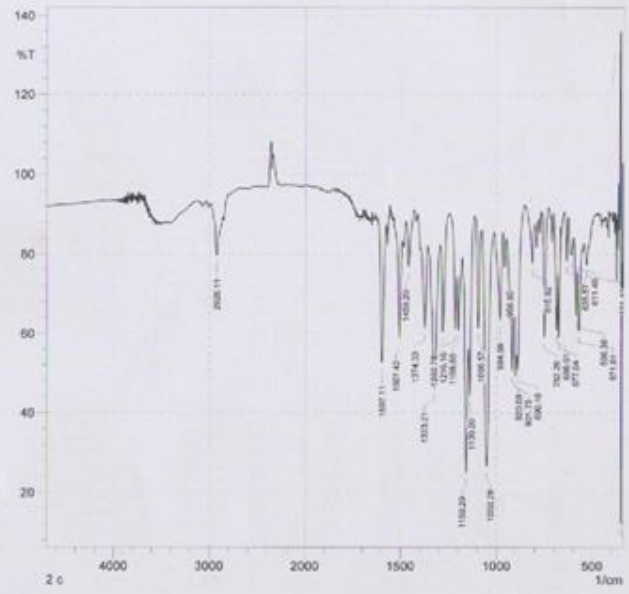
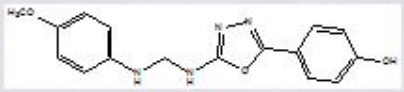


	Peak	Intensity	Corr. Inte	Base (H)	Base (L)	Area	Corr. Are
1	349.13	12.43	52.08	350.09	345.27	2.18	1.46
2	531.41	77.32	6.65	542.98	504.4	3.33	0.62
3	571.91	60.71	16.74	579.63	553.59	3.88	1.17
4	586.38	64.47	13.09	598.92	580.59	2.46	0.65
5	611.45	79.76	0.79	613.38	599.88	1.16	0.02
6	635.57	78.36	11.35	648.1	626.81	1.3	0.4
7	677.04	59.15	19.53	681.86	668.36	2.17	0.91
8	688.61	60.89	18	705.01	682.82	2.83	0.91
9	752.26	59.01	32.1	766.73	730.08	3.38	1.92
10	815.92	77.72	11.35	845.81	800.49	3.06	0.91
11	890.18	80.84	8.36	895	855.46	4.73	0.36
12	901.75	49.27	9.85	910.43	895.96	3.81	0.56
13	920.08	50.58	19.83	949.01	911.4	6.61	1.78
14	958.65	76.71	7.94	968.3	949.97	1.7	0.37
15	984.69	63.45	21.92	1013.63	969.26	5.4	2.35
16	1000.28	26.66	59.21	1079.21	1014.59	15.28	10.97
17	1096.57	61.36	26.34	1114.89	1080.17	3.7	1.73
18	1139	44.43	18.19	1145.75	1115.86	4.89	1.01
19	1158.29	24.89	38.97	1179.51	1146.72	10.08	4.15
20	1198.8	60.8	13.53	1206.51	1189.15	2.93	0.7
21	1216.16	61.12	16.61	1247.02	1207.48	4.13	0.9
22	1260.78	60.31	24.18	1293.31	1247.99	5.05	2.17
23	1323.21	46.12	36.09	1359.86	1294.28	11.57	6.08
24	1374.33	61.31	20.36	1393.62	1360.82	4.84	1.95
25	1459.2	76.62	11.53	1470.77	1438.84	2.56	0.86
26	1507.42	59.06	25.56	1532.5	1492.99	4.72	1.94
27	1597.11	52.74	34.29	1615.44	1578.79	5.64	3.43
28	2926.11	79.63	11.48	2964.94	2862.46	7.85	2.64



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User; Administrator

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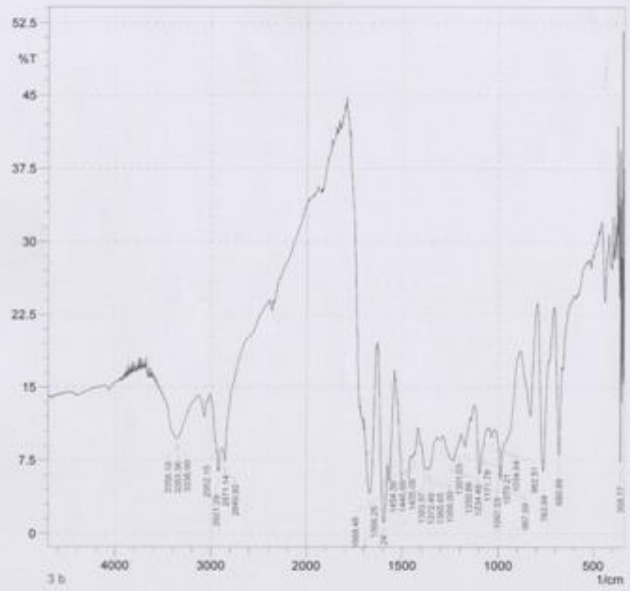
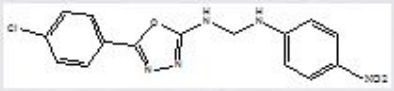


	Peak	Intensity	Corr. Inte	Base (H)	Base (L)	Area	Corr. Are
1	358.77	7.307	24.519	360.7	357.81	2.21	0.757
2	680.89	8.052	11.533	705.97	664.5	35.793	6.737
3	763.84	6.394	14.183	794.7	735.87	53.09	12.554
4	962.51	9.315	0.138	963.48	883.43	70.229	1.169
5	987.59	5.55	4.419	1011.7	964.44	50.912	3.547
6	1034.84	9.897	0.81	1046.42	1024.24	21.892	0.368
7	1079.21	8.565	0.252	1082.1	1055.1	27.2	0.061
8	1097.53	6.11	4.042	1125.5	1083.07	44.176	3.059
9	1171.79	8.912	1.997	1192.05	1144.79	47.175	1.902
10	1234.48	7.434	0.791	1245.09	1193.01	55.759	1.014
11	1250.88	7.692	0.305	1287.53	1246.06	44.154	0.375
12	1301.03	9.587	0.168	1305.85	1288.49	17.561	0.09
13	1356	6.703	0.084	1356.97	1320.32	39.377	0.078
14	1365.65	6.591	0.09	1368.54	1363.72	5.685	0.018
15	1372.4	6.553	0.094	1374.33	1369.5	5.701	0.018
16	1383.97	6.644	0.621	1395.54	1380.11	17.586	0.369
17	1435.09	8.02	0.568	1437.02	1419.66	18.103	0.388
18	1446.66	7.839	0.124	1448.59	1441.84	7.42	0.015
19	1454.38	7.762	0.132	1457.27	1452.45	5.341	0.024
20	1485.24	2.24	5.671	1507.42	1458.23	67.816	13.613
21	1596.25	5.227	4.472	1575.89	1540.21	37.367	3.207
22	1595.18	1.159	10.45	1624.12	1576.86	64.985	20.349
23	1668.48	4.118	0.604	1669.45	1635.69	36.012	1.335
24	2849.92	7.439	1.744	2866.32	2610.74	208.51	0.917
25	2871.14	8.38	0.147	2879.82	2867.28	13.427	0.05
26	2921.29	6.351	2.765	2947.33	2885.6	68.182	3.804
27	2952.15	9.35	0.187	3010.98	2950.22	55.535	0.094
28	3336	9.885	0.033	3336.96	3277.17	58.222	0.135
29	3353.36	9.731	0.044	3355.29	3336.96	18.49	0.029
30	3358.18	9.726	0.036	3362.04	3356.25	5.853	0.006



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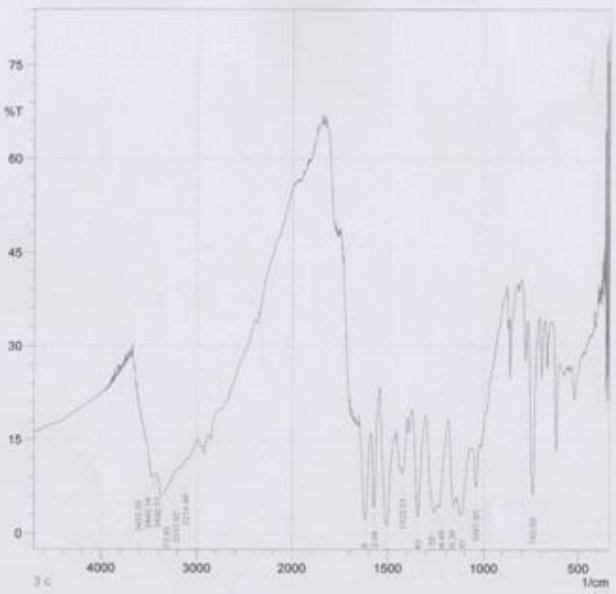
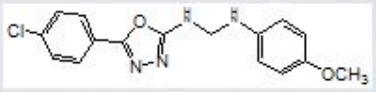


	Peak	Intensity	Corr. Inte	Base (H)	Base (L)	Area	Corr. Are
1	740.69	6.338	27.984	769.62	707.9	43.243	14.589
2	1041.6	7.5	6.538	1063.78	1023.27	38.868	4.303
3	1122.61	2.977	5.14	1142.86	1075.35	82.365	11.507
4	1156.36	4.382	5.32	1184.33	1143.83	45.056	5.092
5	1234.48	4.313	1.755	1240.27	1185.3	56.098	1.281
6	1258.59	3.369	5.327	1301.99	1241.23	72.342	9.648
7	1344.43	2.801	16.559	1374.33	1302.96	70.492	19.484
8	1423.51	9.413	2.809	1430.26	1399.4	28.199	1.866
9	1505.49	1.398	17.225	1540.21	1472.7	80.699	30.738
10	1572.04	4.22	14.425	1587.47	1546	39.309	9.691
11	1619.29	2.213	14.638	1648.23	1588.43	66.728	20.392
12	3214.48	9.971	0.021	3215.44	3198.08	17.231	0.001
13	3310.92	7.228	0.028	3311.89	3217.37	100.943	0.097
14	3373.61	4.974	3.633	3416.05	3312.85	120.09	8.572
15	3430.51	9.643	0.009	3431.48	3426.66	4.895	0.001
16	3440.16	9.477	0.02	3441.12	3436.3	4.918	0
17	3456.55	9.027	0.072	3458.48	3447.87	11.01	0.021



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Apodization; Happ-Genzel
User; Administrator

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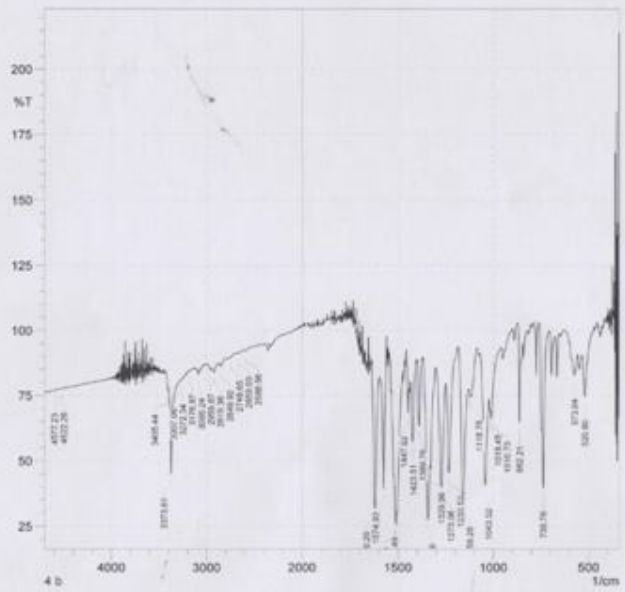
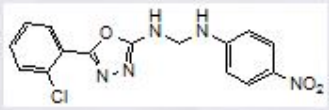


	Peak	Intensity	Corr. Inte	Base (H)	Base (L)	Area	Corr. Are
1	520.8	74.59	17.85	534.3	493.79	2.82	1.62
2	573.84	82.71	9.37	596.99	558.41	2.35	1.03
3	738.76	39.63	28.34	744.55	706.93	3.91	1.24
4	862.21	64.68	35.02	873.78	844.85	1.56	1.4
5	1010.73	66.39	5.5	1015.56	960.58	4.27	0.22
6	1018.45	68.96	2.07	1023.27	1016.52	1.03	0.05
7	1043.52	40.79	40.13	1089.56	1024.24	9.25	5.25
8	1118.75	74.47	6.5	1128.39	1084.99	3.7	0.71
9	1159.26	33.8	52.4	1185.3	1129.36	11.7	7.78
10	1233.52	45.57	37.8	1249.91	1186.26	8.5	4.52
11	1273.06	40.01	42.37	1301.03	1250.88	10.34	6.18
12	1329.96	46.87	3.82	1331.89	1301.99	4.62	0.11
13	1343.46	27.36	32.81	1389.5	1332.86	10.98	4.24
14	1389.76	63.28	25.64	1389.4	1375.29	2.87	1.65
15	1423.51	57.66	24.6	1433.16	1406.15	4.04	1.93
16	1447.62	68.43	13.89	1456.23	1442.8	1.63	0.49
17	1505.49	29.75	3.98	1507.42	1477.52	7.79	0.14
18	1511.28	25.78	8.19	1518.03	1507.42	5.8	0.9
19	1574.93	39.34	49.28	1595.18	1560.46	5.97	3.99
20	1620.26	31.97	47.26	1632.8	1596.15	9.11	5.55
21	2588.56	92.3	0.04	2589.52	2550.94	1.23	0
22	2659.93	91.19	0.33	2665.71	2622.31	1.59	0.02
23	2748.65	89.75	0.04	2749.62	2699.47	2.15	0
24	2849.92	86.09	1.86	2868.24	2811.34	3.29	0.21
25	2919.36	83.86	2.27	2937.68	2877.89	4	0.24
26	2959.87	84.72	1.28	2975.3	2941.54	2.33	0.11
27	3085.24	83.03	2.06	3095.85	3067.88	2.11	0.15
28	3176.87	84.54	0.26	3177.83	3157.58	1.43	0
29	3272.34	80.74	0.22	3273.31	3260.77	1.14	0.01
30	3307.06	79.02	0.36	3308.99	3298.38	1.06	0.01
31	3373.61	45.1	32.76	3397.72	3313.82	13.93	4.97
32	3405.44	76.74	1.53	3431.48	3401.58	2.84	0.08
33	4522.26	77.53	0.1	4524.19	4514.54	1.06	0
34	4577.23	77.19	0.02	4578.2	4568.55	1.08	0



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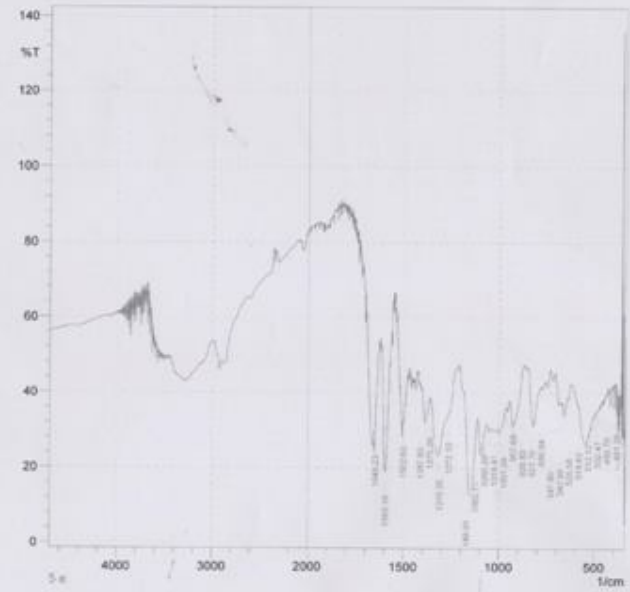
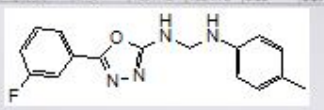


	Peak	Intensity	Corr. Inte	Base (H)	Base (L)	Area	Corr. Are
1	343.34	5.24	30.69	344.31	342.45	2.62	1.23
2	347.2	21.6	10.57	348.16	345.27	1.68	0.22
3	352.99	27.57	17.28	353.95	349.13	1.9	0.6
4	369.38	30.09	9.14	371.31	364.56	3.16	0.42
5	375.17	19.83	19.01	377.1	372.27	2.45	0.48
6	383.85	34.14	7.4	386.74	381.92	2.04	0.2
7	481.26	35.5	0.19	482.22	475.47	3	0.01
8	493.79	34.54	0.29	495.72	488.97	3.1	0.02
9	502.47	33.5	0.52	503.44	496.62	2.25	0.02
10	512.12	32.62	0.2	513.08	504.4	4.14	0.01
11	519.83	30.47	0.41	520.8	513.08	3.86	0.02
12	526.58	28.85	0.76	528.51	521.76	3.56	0.02
13	542.98	26.15	0.24	543.94	529.48	8.07	0.03
14	547.8	25.44	1.78	582.52	543.94	20.62	0.89
15	660.64	33.94	3.65	667.39	638.46	12.81	0.69
16	821.7	31.2	13.59	850.64	782.16	29.18	4.93
17	926.83	31.16	9.33	946.12	872.82	31.93	4.72
18	957.69	35.13	1.81	965.4	947.08	8.08	0.2
19	1001.09	29.18	2.15	1008.8	966.37	21.28	0.92
20	1019.41	29.84	0.49	1037.74	1009.77	14.58	0.09
21	1050.28	29.67	1.26	1064.74	1038.7	13.48	0.22
22	1092.71	22.76	9.76	1112	1065.71	25.86	3.2
23	1149.61	12.52	24.43	1186.26	1112.96	48.11	16.21
24	1272.1	33.01	0.3	1273.06	1227.73	18.81	0.46
25	1319.35	23.18	5.55	1337.68	1274.03	35.88	2.53
26	1375.29	34.33	0.64	1376.26	1365.65	4.67	0.03
27	1387.83	31.66	5.7	1403.26	1377.22	11.9	0.88
28	1502.6	31.82	1.52	1504.53	1492.95	5.45	0.18
29	1595.18	18.95	26.37	1615.44	1578.79	20.01	7.43
30	1648.23	29.58	3.76	1651.12	1634.73	6.99	0.27



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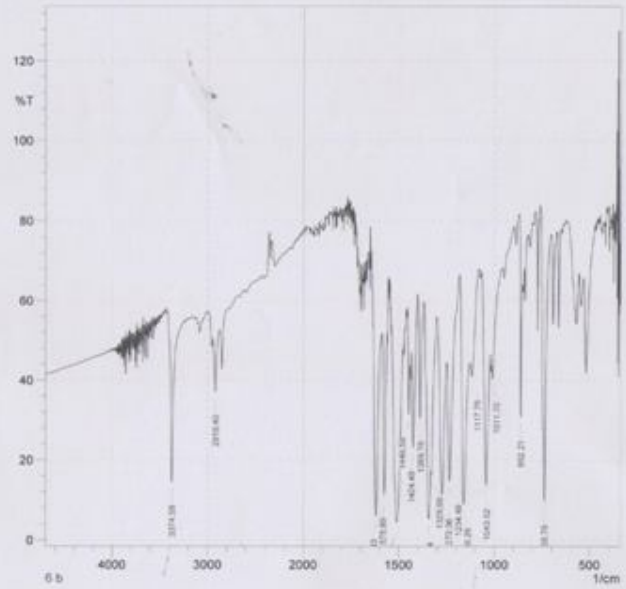
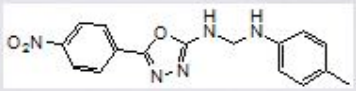


	Peak	Intensity	Corr. Inte	Base (H)	Base (L)	Area	Corr. Are
1	738.76	9.64	25.49	744.55	705.97	13.54	3.01
2	862.21	30.81	40.7	872.82	854.49	4.42	1.84
3	1011.7	40.31	5.15	1015.56	962.51	13.03	0.37
4	1043.52	13.69	42.34	1067.64	1023.27	22.02	10.76
5	1117.79	40.87	9.19	1128.39	1084.03	12.78	1.43
6	1159.26	8.93	48.01	1182.4	1129.36	29.52	15.42
7	1234.48	14.74	33.62	1248.95	1202.66	22.67	8.91
8	1273.06	11.32	38.33	1301.03	1249.91	28.32	12.82
9	1329.96	16.52	1.62	1330.93	1301.99	12.17	0.07
10	1341.54	5.4	23.12	1367.58	1331.89	26.69	8.73
11	1389.76	30.55	30.26	1402.3	1371.43	10.24	3.52
12	1424.48	23.11	28.24	1436.09	1407.12	12	4.19
13	1448.59	31.46	18.13	1455.34	1443.77	4.38	0.86
14	1507.42	4.63	20.11	1519.96	1478.49	35.09	11.48
15	1575.89	11.44	40.42	1588.43	1562.39	14.04	6.62
16	1618.33	6.21	55.78	1644.37	1589.4	29.02	17.03
17	2918.4	37.07	14.32	2944.44	2878.85	22.34	3.5
18	3374.56	14.51	36.62	3401.58	3295.49	42.79	12.27



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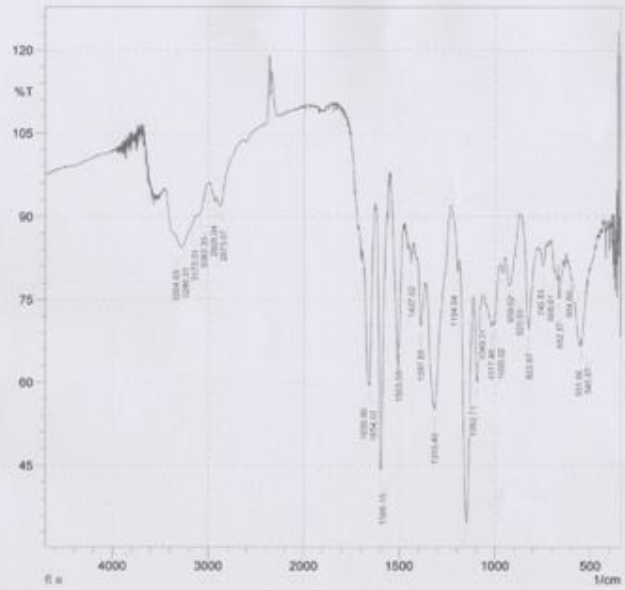
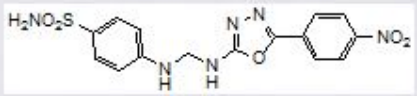


	Peak	Intensity	Corr. Inte	Base (H)	Base (L)	Area	Corr. Are
1	545.87	66.939	0.84	586.84	528.51	2.786	0.038
2	551.66	66.497	0.696	554.55	547.8	1.175	0.013
3	634.8	80.81	1.93	641.35	627.85	1.179	0.071
4	662.57	75.168	6.322	668.36	642.32	2.738	0.454
5	688.61	80.173	0.761	705.97	686.68	1.59	0.036
6	745.51	81.141	3.545	763.84	732.01	2.62	0.314
7	822.67	69.587	18.006	860.28	777.34	8.331	3.416
8	923.93	77.486	7.698	948.04	872.82	6.447	1.733
9	959.62	79.844	2.202	971.19	949.01	2.04	0.133
10	1003.02	70.44	2.017	1007.84	972.16	4.209	0.119
11	1017.48	70.222	1.608	1043.52	1008.8	5.024	0.183
12	1049.31	73.908	0.676	1063.78	1044.49	2.441	0.034
13	1092.71	60.049	15.657	1112.96	1064.74	7.722	1.905
14	1149.61	34.717	44.1	1185.3	1113.93	18.791	11.368
15	1194.94	79.996	4.139	1229.66	1186.26	2.837	0.239
16	1316.46	55.029	27.578	1359.86	1230.63	19.522	10.2
17	1367.83	70.1	10.452	1403.26	1365.65	4.728	1.104
18	1437.02	81.635	3.233	1446.66	1426.41	1.563	0.124
19	1503.56	66.287	0.848	1504.53	1479.45	2.708	0.042
20	1596.15	44.193	44.558	1622.19	1572.04	8.895	6.297
21	1654.01	59.209	3.501	1655.94	1634.73	2.768	0.101
22	1659.8	59.55	2.554	1682.95	1656.91	4.636	0.456
23	2873.07	91.802	3.379	2910.68	2762.16	2.922	0.895
24	2928.04	92.641	0.985	2949.26	2911.64	1.155	0.084
25	3082.35	90.549	0.143	3084.28	3014.84	2.202	0.066
26	3173.01	88.222	0.046	3173.97	3131.54	2.125	0.014
27	3246.31	85.085	0.064	3247.27	3180.72	4.1	0.001
28	3264.63	84.524	0.085	3266.56	3247.27	1.382	0.006



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User; Administrator

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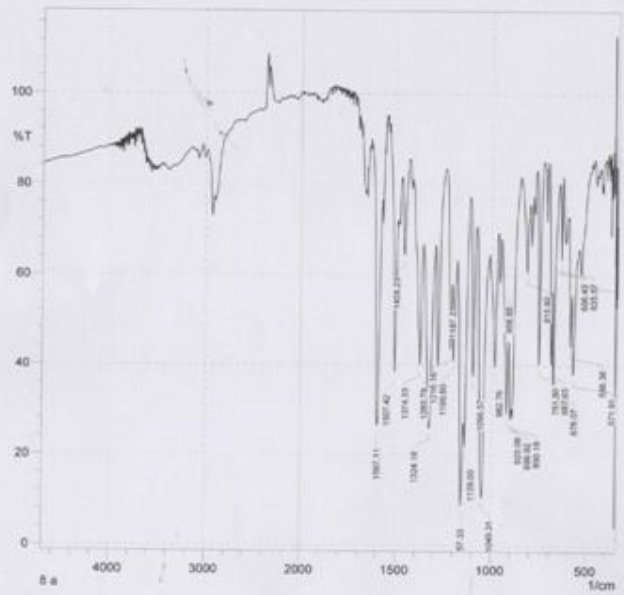
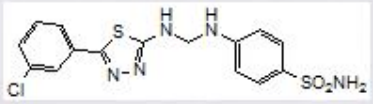


	Peak	Intensity	Corr. Inte	Base (H)	Base (L)	Area	Corr. Are
1	350.09	4.2	54.73	351.06	346.23	2.2	1.45
2	531.41	60.36	9.06	542.98	488.01	8.14	0.99
3	571.91	38.08	19.81	579.63	552.62	8.14	2.07
4	586.38	44.27	16.61	598.92	580.59	4.66	1.01
5	635.57	63.61	17.7	645.21	626.81	2.19	0.7
6	666.43	63.47	1.88	667.39	646.17	2.82	0.03
7	676.07	35.78	23.47	681.86	668.36	4.55	1.55
8	687.65	40.34	22.24	705.01	682.82	4.94	1.4
9	751.3	40.32	43.75	766.73	729.12	6.4	3.6
10	815.92	60.71	17.02	846.71	800.49	6.8	1.78
11	890.18	28.48	7.44	895	849.67	10.51	0.43
12	899.82	28.1	6.17	910.43	895.96	5.93	0.62
13	920.08	29.3	21.03	949.01	911.4	12.73	2.99
14	958.65	56.39	10.41	967.33	949.97	3.43	0.6
15	982.76	39.74	27.95	1011.7	968.3	11.66	4.09
16	1049.31	10.8	57.21	1079.21	1012.86	29.54	18.21
17	1096.57	37.63	36.43	1112.96	1080.17	7.87	3.56
18	1139	22.05	14.6	1144.79	1113.93	10.16	1.17
19	1157.33	8.98	30.42	1179.51	1145.75	19.85	6.75
20	1187.23	58.23	0.68	1188.19	1180.47	1.68	0.03
21	1198.8	40.97	17.05	1206.51	1188.19	5.59	1.26
22	1216.16	43.57	21.34	1239.31	1207.48	6.54	1.61
23	1280.78	40.1	30.55	1293.31	1240.27	10.22	3.68
24	1324.18	26.02	40.75	1358.9	1294.28	22.41	11.08
25	1374.33	40.13	30.67	1413.87	1359.86	12.45	5.27
26	1458.23	64.6	17.73	1470.77	1438.94	4.12	1.46
27	1507.42	38.57	40.28	1537.32	1492.95	8.16	4.24
28	1597.11	26.67	54.26	1622.19	1575.89	12.84	8.62



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User; Administrator

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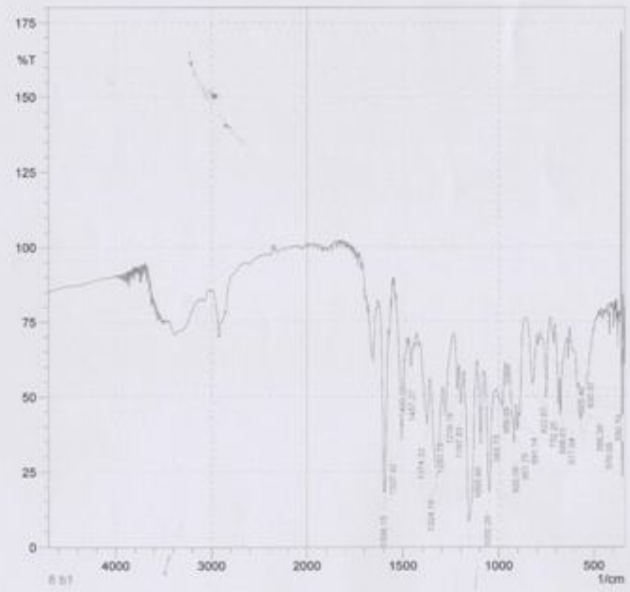

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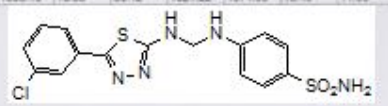
1	343.34	60.99	20.17	347.2	340.45	1.05	0.43
2	351.06	29.05	44.06	352.99	348.16	1.21	0.64
3	353.95	23.14	63.84	357.81	352.99	1	0.93
4	532.37	57.26	1.01	534.3	504.4	5.62	0.04
5	544.91	52.52	0.43	545.87	535.26	2.69	0
6	550.7	51.57	0.68	554.55	547.8	1.92	0.02
7	570.95	42.33	11.09	579.63	558.41	6.95	1.12
8	586.38	48.25	9.35	598.92	580.59	4.77	0.58
9	635.57	62.76	8.61	642.32	628.81	2.32	0.35
10	665.46	61.04	0.85	667.39	643.28	4.46	0.04
11	677.04	43.94	13.45	681.86	667.39	4.19	0.82
12	688.61	47.42	13.15	705.97	682.82	5.3	0.86
13	752.26	49.5	23.75	772.52	732.01	7.55	2.08
14	822.67	54.24	18.96	853.53	799.52	10.33	3.1
15	891.14	43.92	4.09	894.04	868	5.55	0.19
16	901.75	38.86	6.9	909.47	895	5.42	0.51
17	920.08	34.49	16.19	948.04	910.43	12.95	2.69
18	958.65	54.29	6.64	967.33	949.01	4.4	0.45
19	983.73	44.86	9.94	997.23	968.3	8.52	1.02
20	1060.28	18.08	38.11	1075.35	1012.66	25.66	9.61
21	1095.6	34.23	26.25	1112.96	1076.32	11.56	3.52
22	1157.33	8.07	51.76	1181.44	1113.93	38.44	23.62
23	1197.83	47.49	12.14	1207.48	1182.4	6.79	1.11
24	1216.16	52.31	11.15	1235.45	1208.44	5.68	0.73
25	1280.78	42.38	14.25	1291.39	1236.41	13.5	2.03
26	1324.18	23.23	31.29	1358.9	1292.35	28.74	11.22
27	1374.33	40.41	19.05	1413.87	1359.86	14.97	3.7
28	1457.27	59.95	7.05	1469.81	1450.52	3.75	0.44
29	1490.06	61.29	2.14	1491.99	1478.49	2.49	0.06
30	1507.42	35.48	27.27	1518.99	1492.95	8.17	2.88
31	1596.15	18.05	58.12	1621.22	1574.93	16.49	11.05



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User; Administrator

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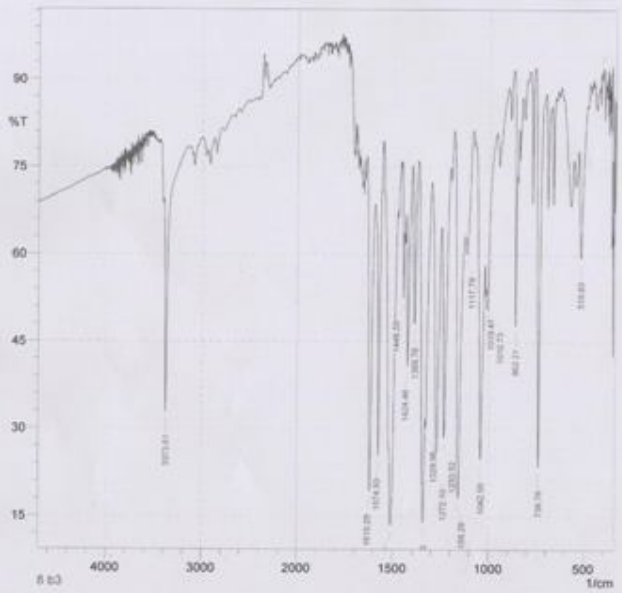

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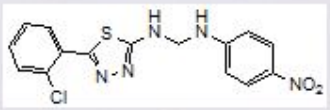
1	519.83	59.441	20.587	532.37	489.94	6.075	2.333
2	738.76	23.556	29.354	744.55	705.01	7.604	1.832
3	862.21	47.831	32.919	872.82	855.46	2.506	1.042
4	1010.73	50.69	6.028	1015.56	983.48	8.419	0.353
5	1019.41	53.088	2.897	1023.27	1016.52	1.774	0.072
6	1042.56	24.801	41.427	1069.56	1024.24	15.324	7.504
7	1117.79	60.153	7.235	1127.43	1084.99	6.946	0.938
8	1158.29	18.089	54.691	1185.3	1128.39	19.714	11.466
9	1233.52	28.467	39.846	1248.95	1204.59	13.216	6.287
10	1272.1	23.703	44.398	1301.03	1249.91	17.132	8.709
11	1329.96	30.198	4.125	1331.89	1301.99	8.101	0.174
12	1342.5	14.082	28.858	1370.47	1332.86	17.64	5.95
13	1389.76	48.026	27.608	1402.3	1371.43	5.999	2.271
14	1424.48	40.731	29.027	1435.09	1403.26	7.616	2.91
15	1448.59	52.604	16.753	1455.34	1442.8	2.578	0.565
16	1511.28	13.538	59.066	1552.75	1478.49	31.455	21.298
17	1574.93	25.479	44.483	1594.22	1562.39	9.448	4.468
18	1619.29	19.316	53.796	1639.55	1595.18	14.971	8.788
19	3373.61	32.82	37.582	3399.65	3279.1	24.515	6.943



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User; Administrator

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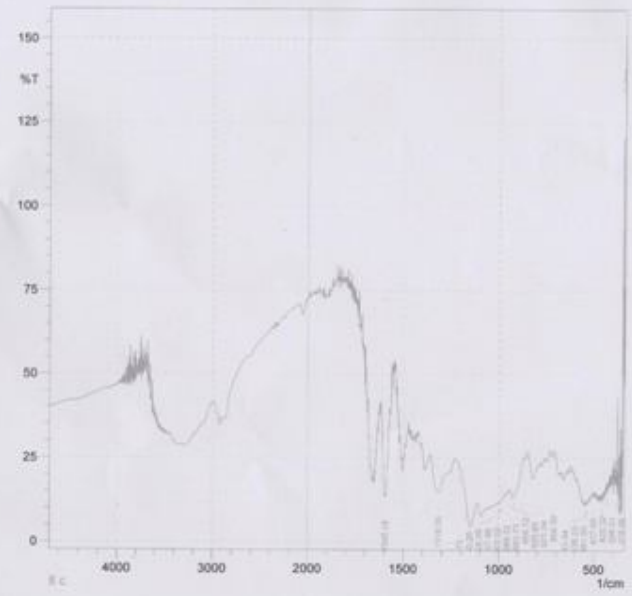
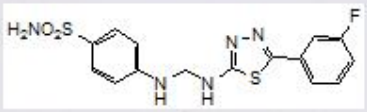


	Peak	Intensity	Corr. Inte	Base (H)	Base (L)	Area	Corr. Are
1	353.95	9.71	24.8	354.91	352.02	2.13	0.97
2	358.77	9.28	17.56	359.74	354.91	3.96	1.24
3	366.49	8.44	13.4	368.42	365.52	2.4	0.48
4	370.34	13.14	9.56	372.27	368.42	2.99	0.55
5	378.06	14.53	15.32	379.99	376.13	2.57	0.59
6	398.31	14.85	4.92	401.21	397.35	2.88	0.19
7	425.32	15.15	2.43	431.1	424.35	5.31	0.19
8	437.86	14.51	2.08	439.78	434.96	3.93	0.19
9	461	12.16	2.31	464.86	459.07	5.05	0.19
10	536.23	12.03	0.22	537.19	531.41	5.24	0.03
11	543.94	10.96	0.86	545.87	541.05	4.5	0.06
12	908.5	15.12	0.24	909.47	869.92	27.91	0.13
13	921.04	13.68	0.24	922	911.4	8.96	0.05
14	925.86	13.29	0.9	943.22	922	18.04	0.42
15	956.72	14.6	0.14	957.69	947.08	8.67	0.04
16	983.73	12.68	0.29	985.66	966.37	16.68	0.11
17	989.52	12.42	0.11	990.48	985.66	4.35	0.02
18	1003.02	11.54	0.23	1003.98	993.37	9.83	0.07
19	1017.48	11.26	0.16	1018.45	1009.77	8.16	0.04
20	1028.09	10.99	0.1	1029.06	1024.24	4.61	0.01
21	1050.28	10.17	0.15	1051.24	1040.63	10.38	0.03
22	1091.75	7.82	3.2	1113.93	1070.53	44.61	3.01
23	1148.65	5.05	0.46	1150.58	1114.89	39.32	0.34
24	1318.39	14.82	7.63	1357.93	1288.49	51.99	7.17
25	1595.18	13.49	21.88	1613.51	1576.86	25.13	8.57



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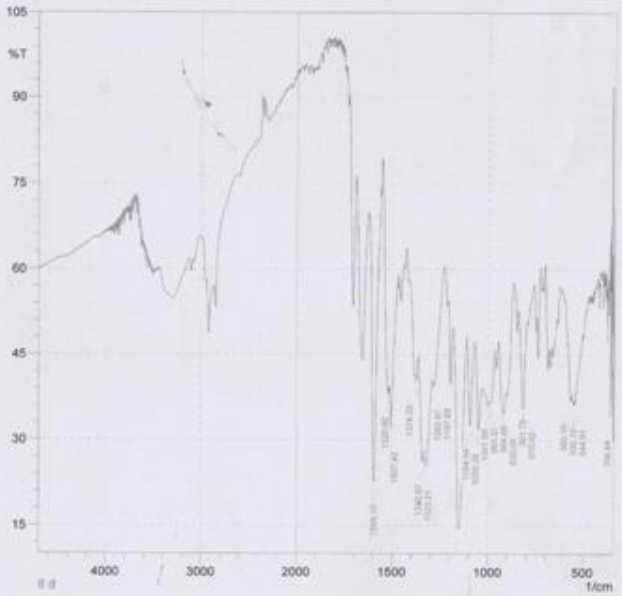
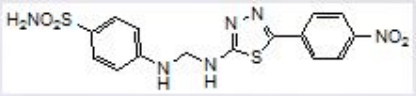


	Peak	Intensity	Corr. Inte	Base (H)	Base (L)	Area	Corr. Are
1	343.34	29.788	38.951	347.2	339.48	2.779	1.444
2	350.84	34.185	45.582	359.74	353.95	1.729	1.196
3	544.91	36.113	1.77	548.77	504.4	18.772	0.269
4	550.7	36.238	0.846	558.41	548.77	4.161	0.036
5	565.16	36.975	3.211	582.52	559.38	9.206	0.509
6	815.92	35.562	15.979	842.92	797.59	16.294	3.306
7	901.75	37.812	3.065	906.57	869.92	12.787	0.908
8	920.08	34.447	6.785	946.12	907.54	16.116	1.767
9	984.69	36.679	0.916	986.62	966.37	8.033	0.238
10	993.37	36.434	0.145	995.3	987.59	3.371	0.008
11	1001.09	36.006	0.884	1030.02	996.27	14.4	0.13
12	1050.28	31.703	11.299	1071.49	1030.99	17.071	2.195
13	1094.64	32.597	15.182	1112.96	1072.46	15.968	2.953
14	1151.54	14.447	34.603	1183.37	1113.93	38.648	17.106
15	1197.83	39.692	12.61	1210.37	1184.33	8.686	1.306
16	1283.67	39.444	3.254	1290.42	1230.63	18.711	0.716
17	1323.21	25.329	6.18	1334.78	1291.39	22.129	1.778
18	1340.57	26.352	0.396	1341.54	1335.75	3.263	0.018
19	1374.33	40.179	2.687	1381.08	1366.61	5.483	0.188
20	1507.42	31.96	9.641	1514.17	1491.99	9.258	1.176
21	1520.92	38.069	1.263	1523.82	1518.99	1.986	0.032
22	1596.15	22.747	43.639	1621.22	1572.04	18.711	9.941



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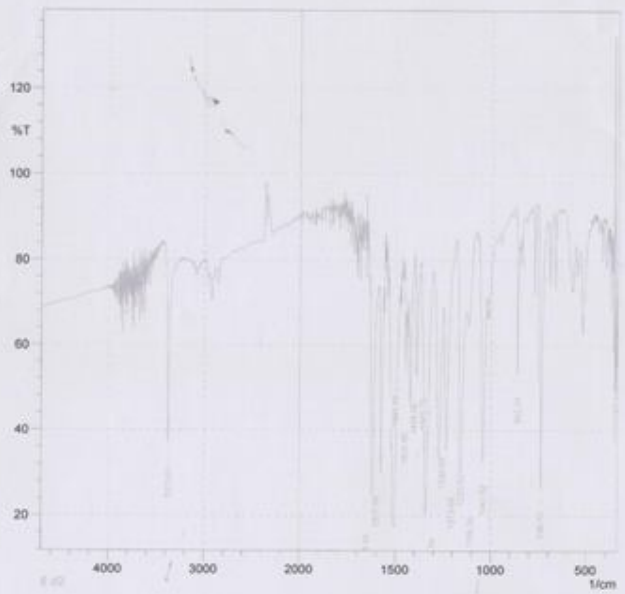
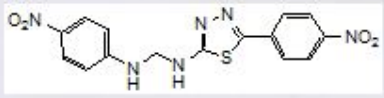


	Peak	Intensity	Corr. Inte	Base (H)	Base (L)	Area	Corr. Are
1	343.34	54.82	18.82	346.23	339.48	1.27	0.4
2	736.76	26.44	31.28	744.55	706.93	6.87	1.86
3	862.21	53.47	34.04	873.78	853.53	2.25	1.13
4	1043.52	32.96	45.02	1067.64	1024.24	10.92	6.31
5	1159.26	25.24	52.36	1182.4	1129.36	15.37	9.01
6	1233.52	35.27	37.76	1249.91	1183.37	13.24	5.61
7	1273.06	29.85	42.89	1301.99	1250.88	14.4	7.44
8	1329.96	39.14	3.61	1331.89	1302.96	5.91	0.09
9	1341.54	20.31	29.32	1367.58	1332.86	13.62	4.77
10	1389.76	52.97	26.12	1403.26	1377.22	4.44	1.78
11	1419.66	51.61	1.97	1420.62	1407.12	2.14	0.01
12	1424.48	44.78	10.38	1435.09	1421.58	3.57	0.5
13	1448.59	53.72	18.61	1455.34	1443.77	2.18	0.57
14	1507.42	17.59	30.34	1519.96	1479.45	18.45	7
15	1575.89	30.13	43.66	1588.43	1563.36	7.15	3.84
16	1618.33	21.23	52.5	1633.76	1593.25	13.32	7.94
17	3373.61	36.85	41.96	3417.98	3338.89	14.62	6.6



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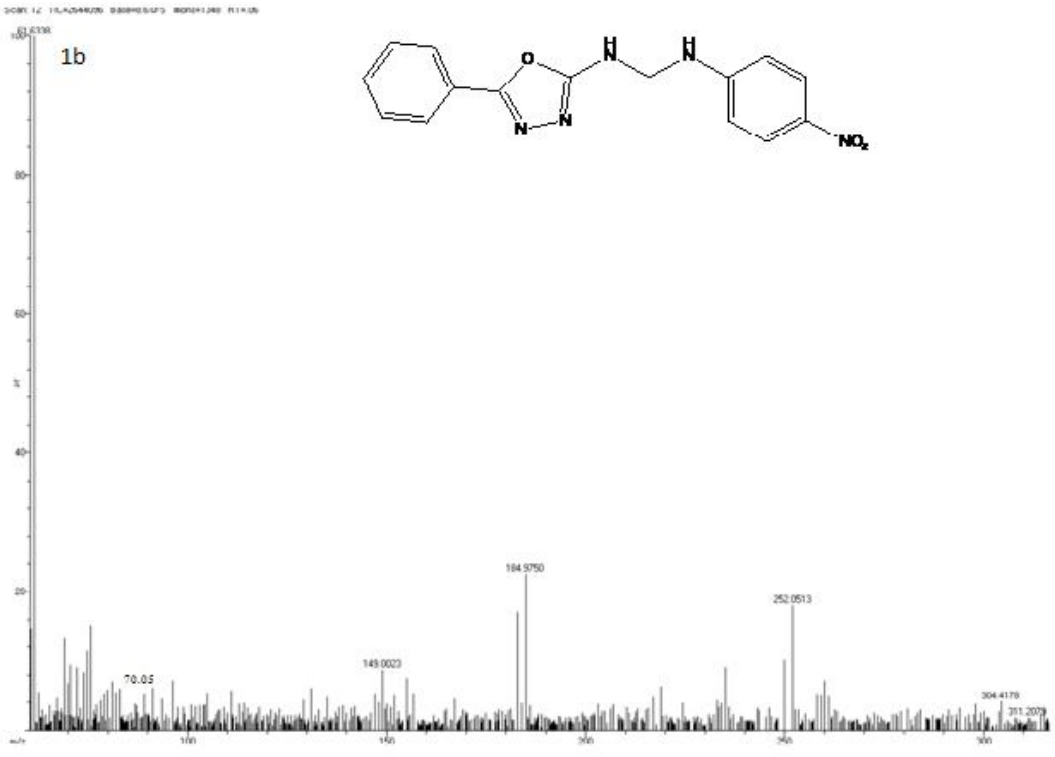
User; Administrator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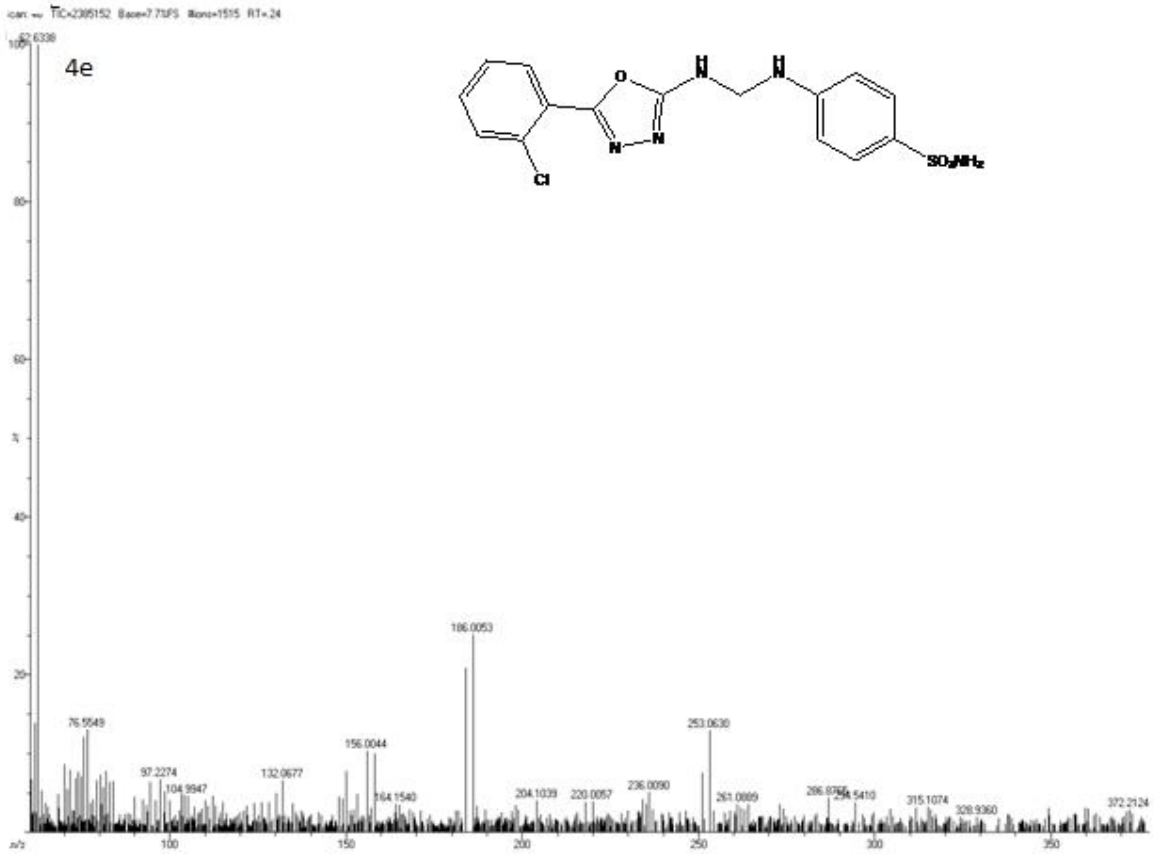
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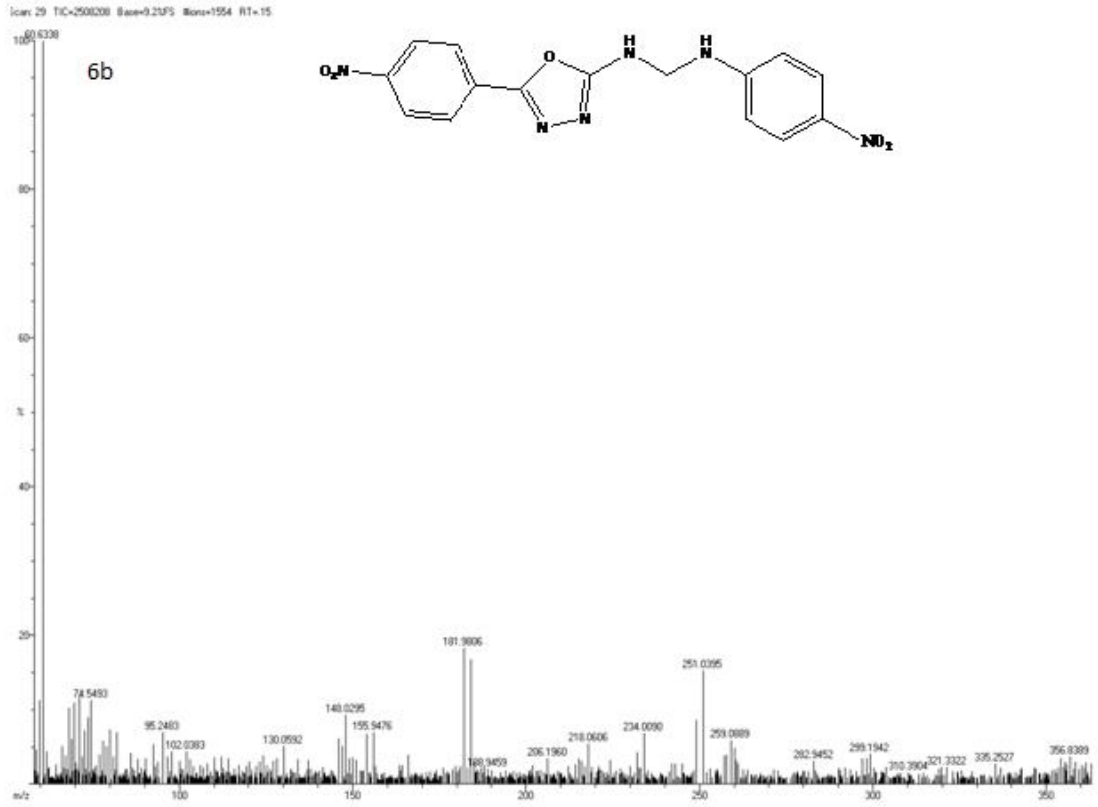
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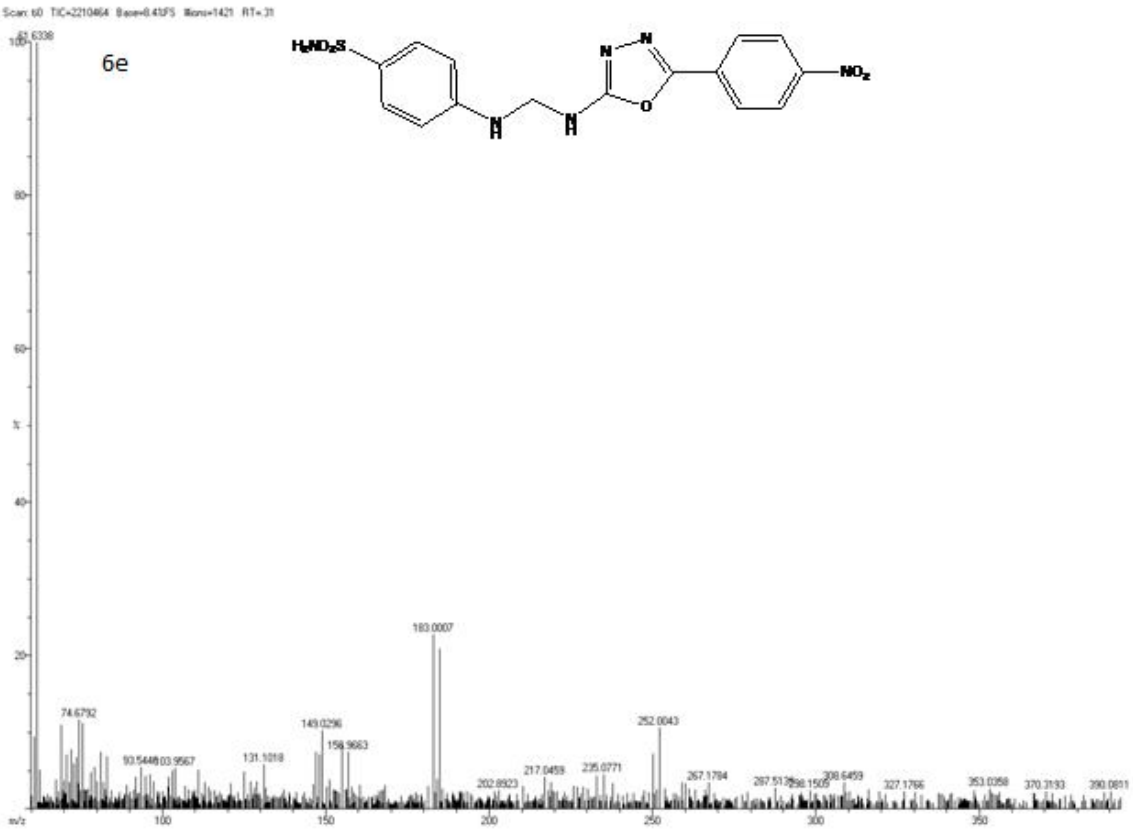
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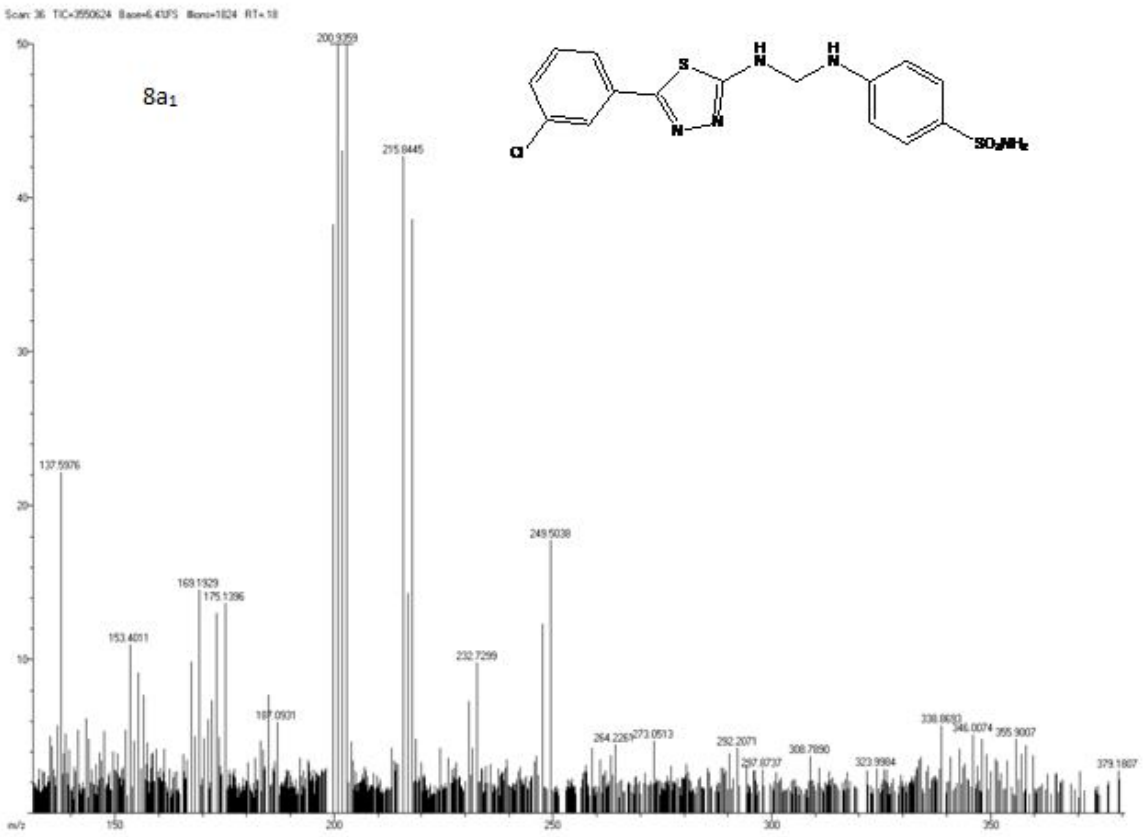
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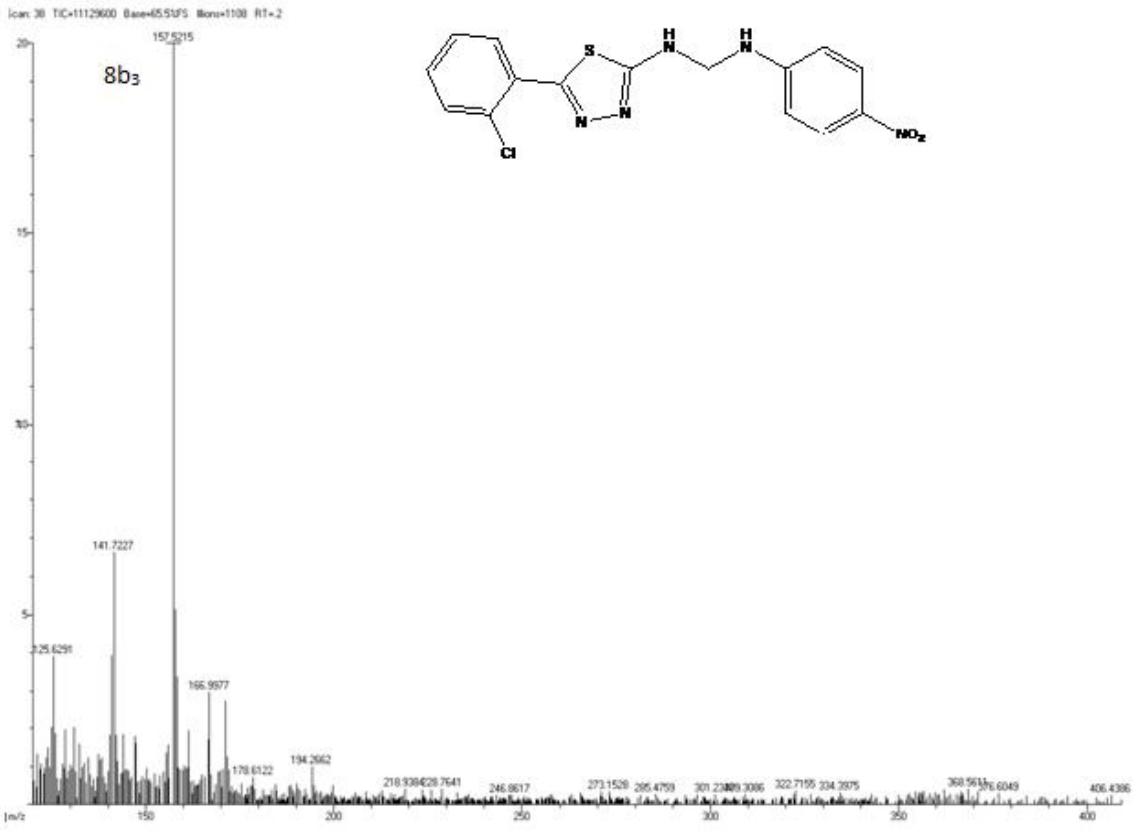
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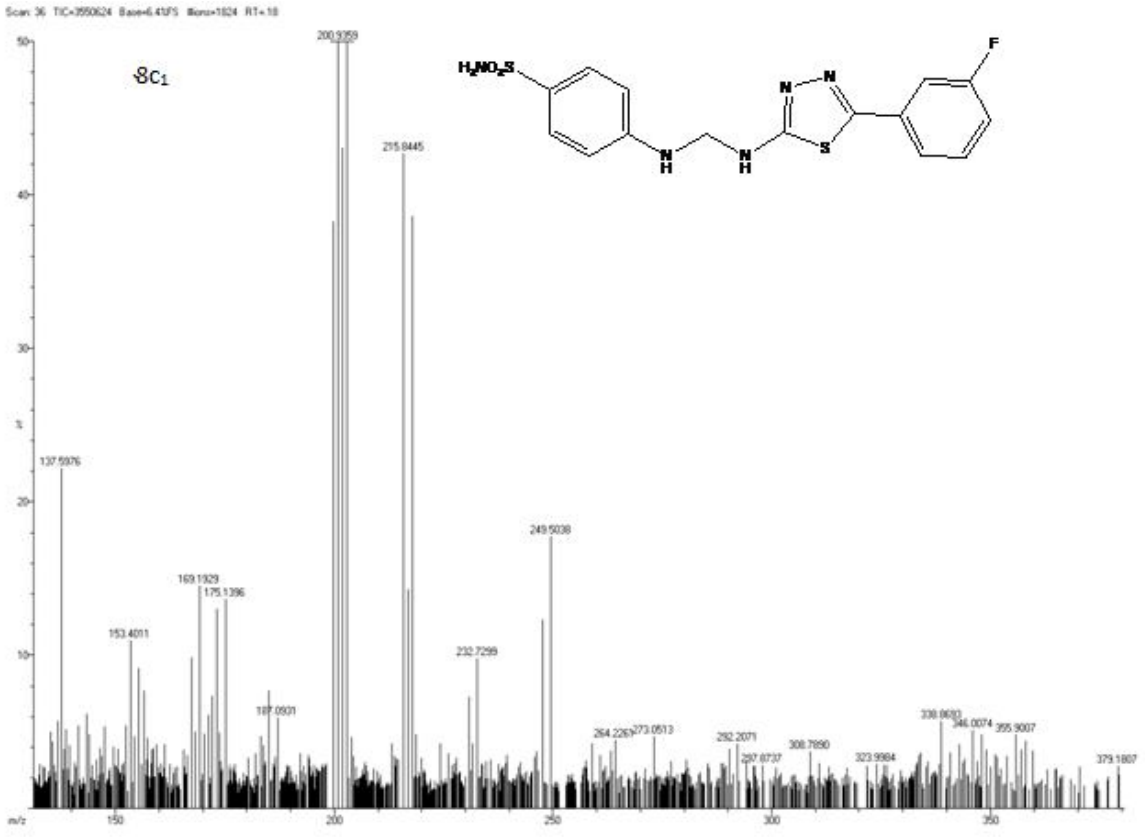
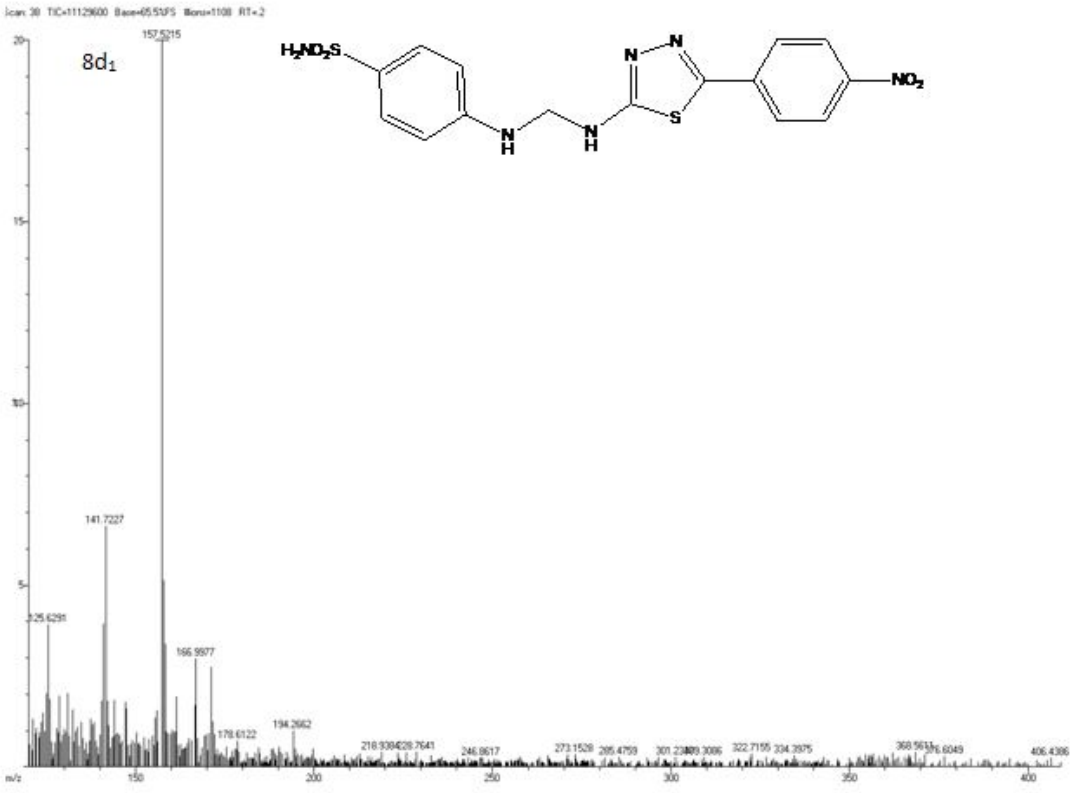
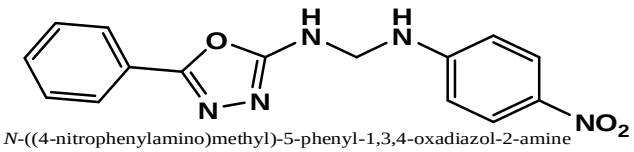
225.6291
166.9977
178.6122
194.2662
218.3384
228.7641
246.8617
273.1528
285.4759
301.2389
309.3086
322.7155
334.3975
368.5613
376.6049
406.4386
m/z
150
200
250
300
350
400
Cl
NO2

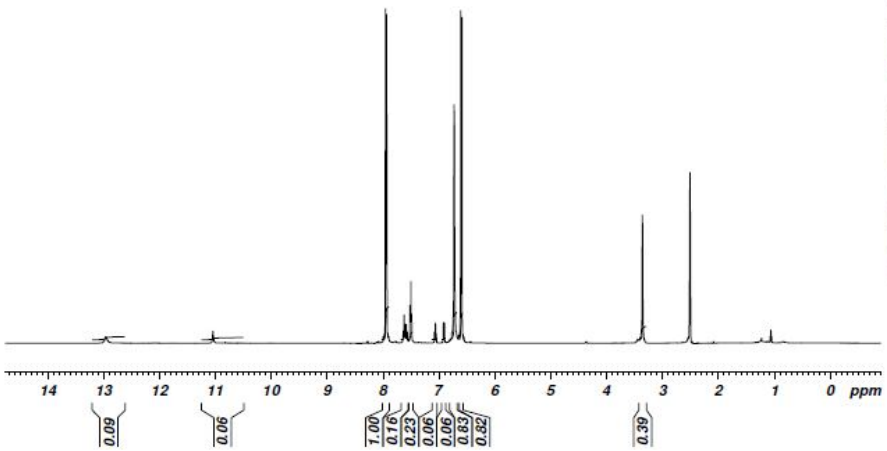

8c1
F
N
N
S
N
H
N
H
200.9359
215.8445
137.5976
249.5038
169.1329
175.1396
153.4011
232.7299
187.0501
264.2267
273.0513
292.2071
297.6737
308.7690
323.9984
338.8630
346.0074
355.9007
379.1807
m/z
150
200
250
300
350


8d1
H2NO2S
NO2
N
N
S
NH
NH
157.5215
141.7227
125.6291
166.9977
178.6122
194.2662
218.9394
229.7641
246.8617
273.1529
285.4759
301.2389
309.3086
322.7155
334.2975
368.5616
376.6049
406.4386
m/z
150
200
250
300
350
400



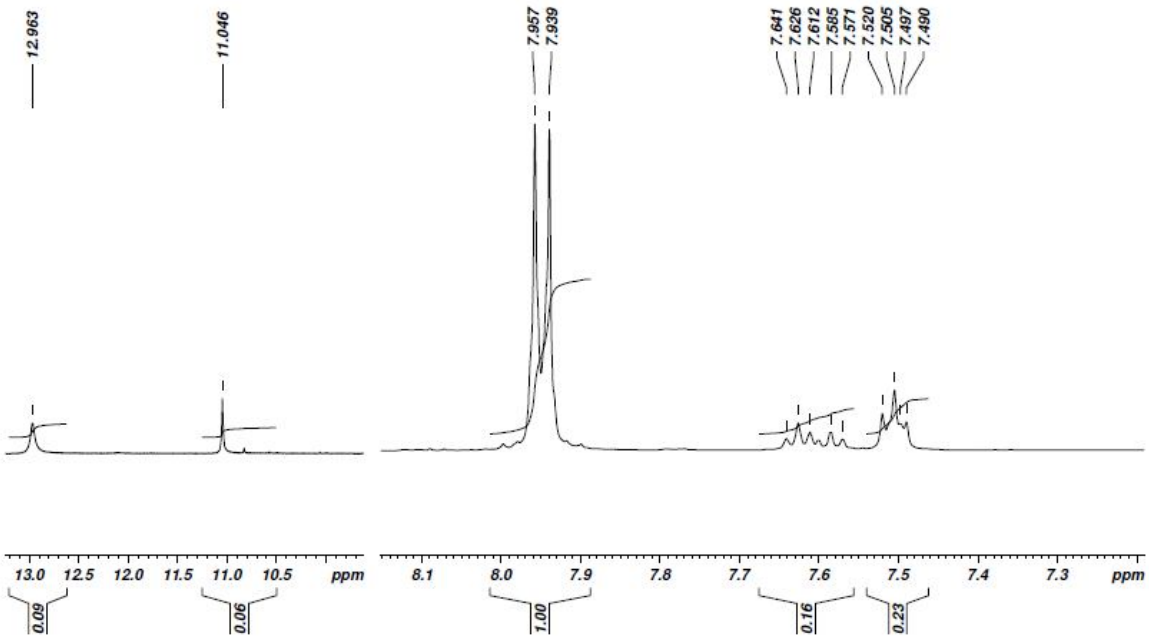
N-((4-nitrophenylamino)methyl)-5-phenyl-1,3,4-oxadiazol-2-amine

Ib.....Smylin.

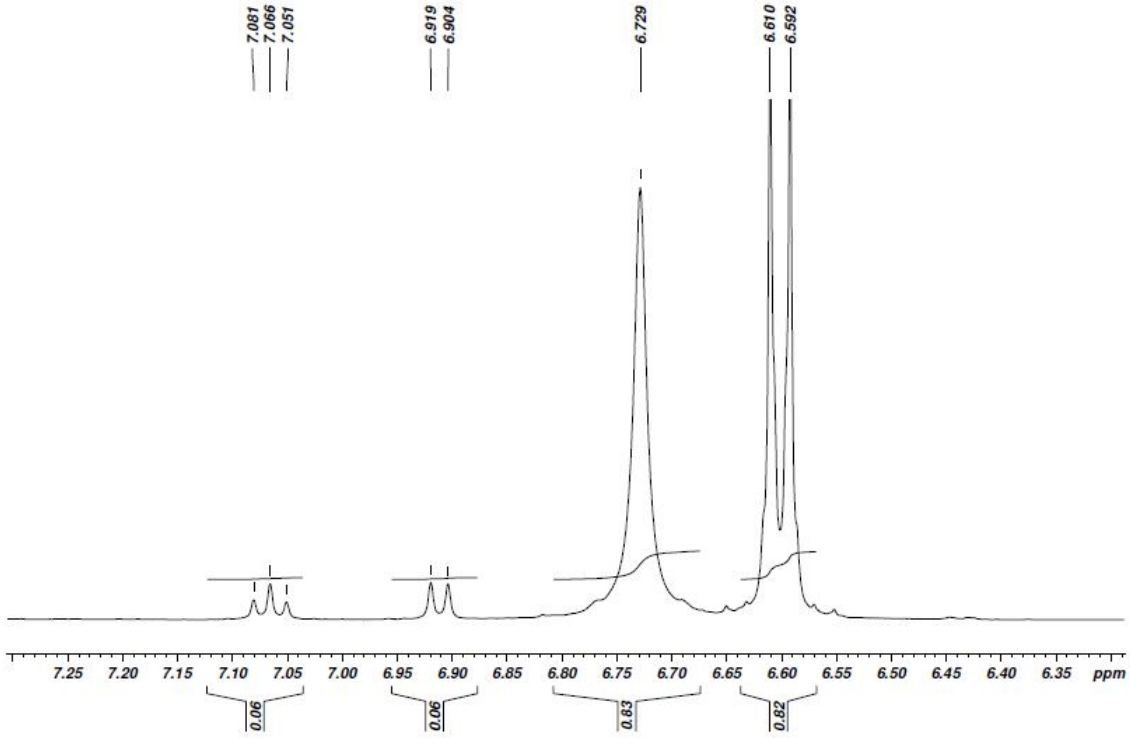


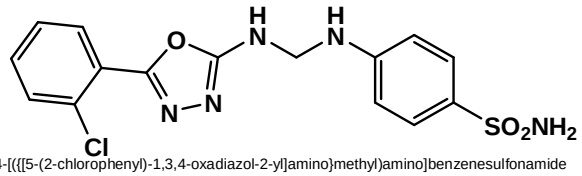

```
Current Data Parameters
NAME          Feb01-2012
EXPNO                 19
PROCNO                 1

F2 - Acquisition Parameters
Date_           20120201
Time               15.55
INSTRUM             spect
PROBHD   5 mm PABBO BB-
PULPROG             zg30
TD                 32768
SOLVENT             DMSO
NS                    32
DS                     2
SWH            10330.578 Hz
FIDRES         0.315264 Hz
AQ             1.5860212 sec
RG                   203
DW                48.400 usec
DE                 6.50 usec
TE                295.5 K
D1             1.00000000 sec
TD0                    1

======== CHANNEL f1 ========
NUC1                  1H
P1                 10.65 usec
PL1                0.00 dB
PL1W         23.53637505 W
SFO1         500.1330885 MHz

F2 - Processing parameters
SI                 32768
SF           500.1300000 MHz
WDW                   EM
SSB                    0
LB                 0.30 Hz
GB                     0
PC                 1.00
```

Ib.....Smylin.


12.963
11.046
7.957
7.939
7.641
7.626
7.612
7.585
7.571
7.520
7.505
7.497
7.490
13.0
12.5
12.0
11.5
11.0
10.5
ppm
8.1
8.0
7.9
7.8
7.7
7.6
7.5
7.4
7.3
ppm
0.09
0.06
1.00
0.16
0.23

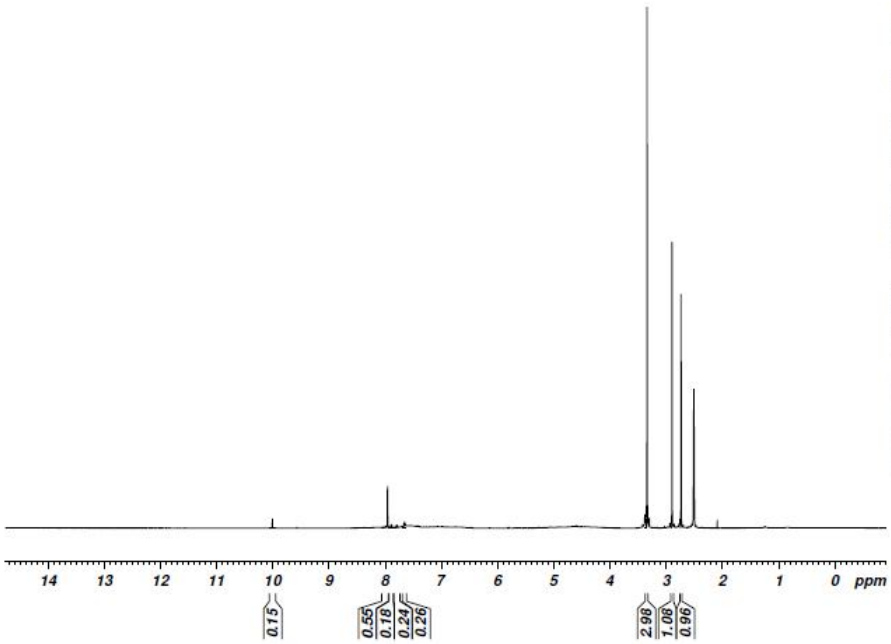
Ib.....Smylin.


7.081
7.066
7.051
6.919
6.904
6.729
6.610
6.592
7.25 7.20 7.15 7.10 7.05 7.00 6.95 6.90 6.85 6.80 6.75 6.70 6.65 6.60 6.55 6.50 6.45 6.40 6.35 ppm
0.06
0.06
0.83
0.82



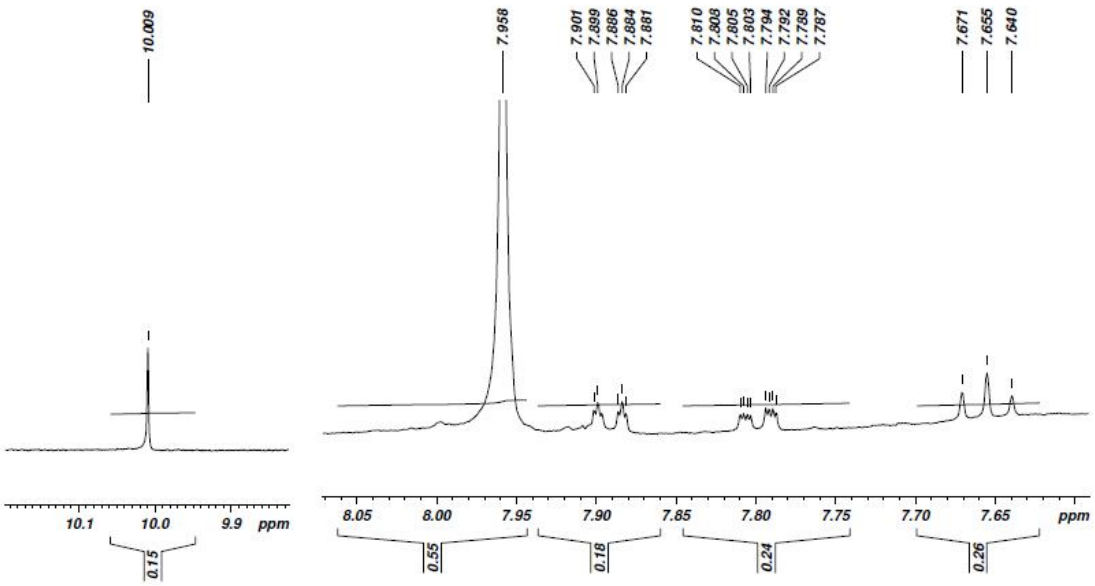
4-[({[5-(2-chlorophenyl)-1,3,4-oxadiazol-2-yl]amino}methyl)amino]benzenesulfonamide

IVe.....Smylin.

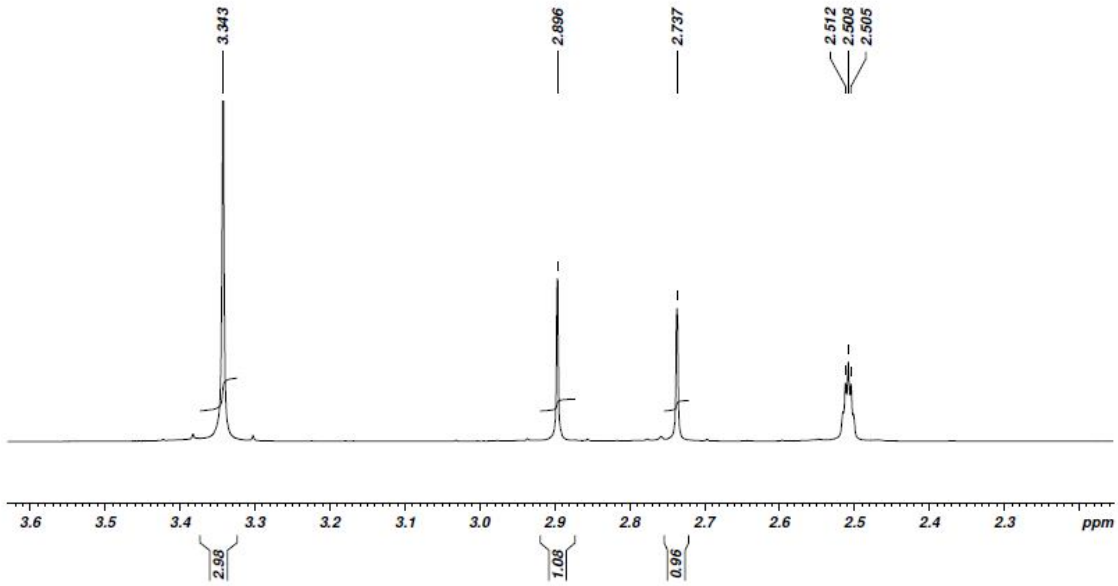



```
Current Data Parameters
NAME          Feb01-2012
EXPNO                 21
PROCNO                 1

F2 - Acquisition Parameters
Date_           20120201
Time               16.04
INSTRUM            spect
PROBHD   5 mm PABBO BB-
PULPROG             zg30
TD                 32768
SOLVENT             DMSO
NS                    32
DS                     2
SWH            10330.578 Hz
FIDRES         0.315264 Hz
AQ             1.5860212 sec
RG                   203
DW                48.400 usec
DE                 6.50 usec
TE                295.5 K
D1             1.00000000 sec
TD0                    1

====== CHANNEL f1 ======
NUC1                  1H
P1                10.65 usec
PL1                0.00 dB
PL1W        23.53637505 W
SFO1        500.1330885 MHz

F2 - Processing parameters
SI                 32768
SF          500.1300000 MHz
WDW                   EM
SSB                    0
LB                 0.30 Hz
GB                     0
PC                 1.00
```

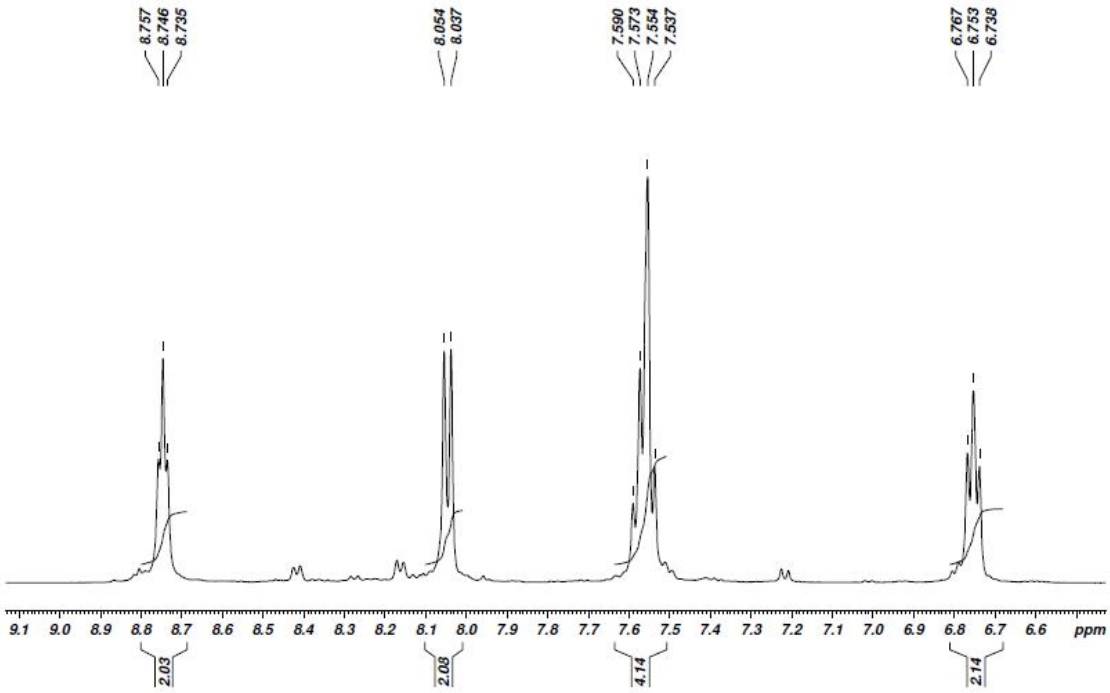
IVe.....Smylin.


10.009
7.958
7.901
7.899
7.886
7.884
7.881
7.810
7.808
7.805
7.803
7.794
7.792
7.789
7.787
7.671
7.655
7.640
10.1
10.0
9.9
ppm
8.05
8.00
7.95
7.90
7.85
7.80
7.75
7.70
7.65
ppm
0.15
0.55
0.18
0.24
0.26

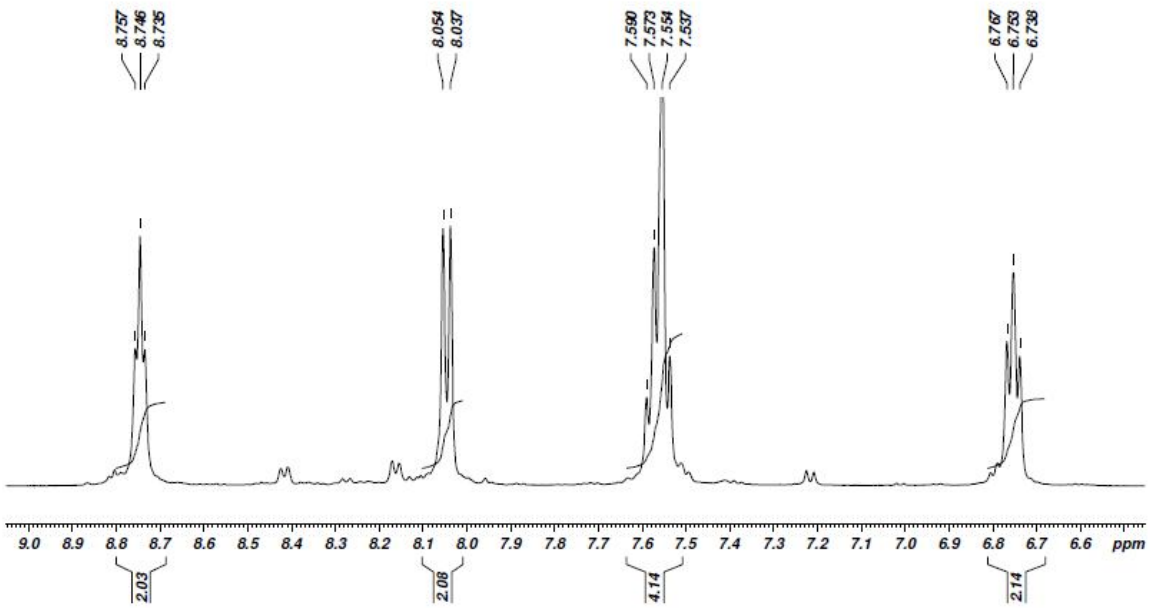
IVe.....Smylin.

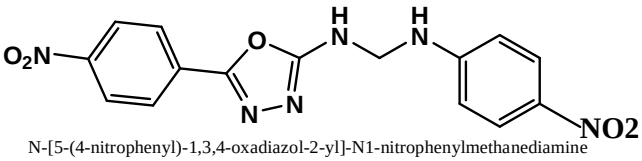

3.343
2.896
2.737
2.512
2.508
2.505
3.6
3.5
3.4
3.3
3.2
3.1
3.0
2.9
2.8
2.7
2.6
2.5
2.4
2.3
ppm
2.98
1.08
0.96

VIb.....Smylin.


8.757
8.746
8.735
8.054
8.037
7.590
7.573
7.554
7.537
6.767
6.753
6.738
9.1 9.0 8.9 8.8 8.7 8.6 8.5 8.4 8.3 8.2 8.1 8.0 7.9 7.8 7.7 7.6 7.5 7.4 7.3 7.2 7.1 7.0 6.9 6.8 6.7 6.6 ppm
2.03
2.08
4.14
2.14

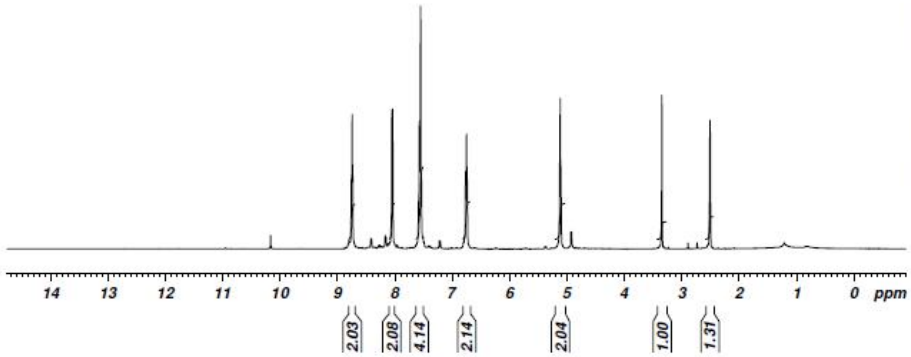
VIb.....Smylin.


8.757
8.746
8.735
8.054
8.037
7.590
7.573
7.554
7.537
6.767
6.753
6.738
9.0 8.9 8.8 8.7 8.6 8.5 8.4 8.3 8.2 8.1 8.0 7.9 7.8 7.7 7.6 7.5 7.4 7.3 7.2 7.1 7.0 6.9 6.8 6.7 6.6 ppm
2.03
2.08
4.14
2.14



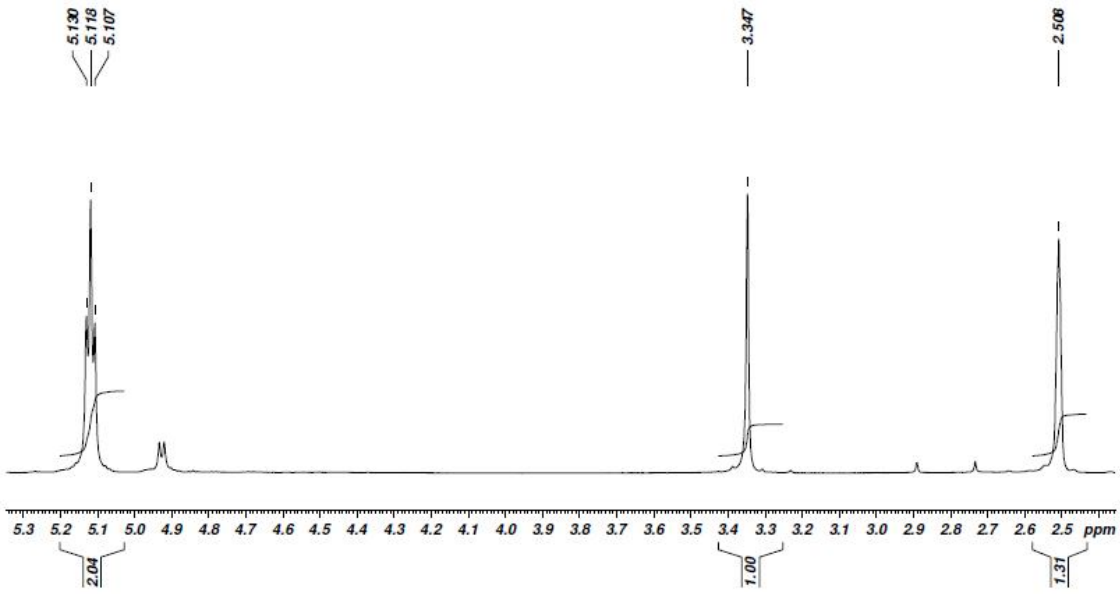
N-[5-(4-nitrophenyl)-1,3,4-oxadiazol-2-yl]-N1-nitrophenylmethanediamine

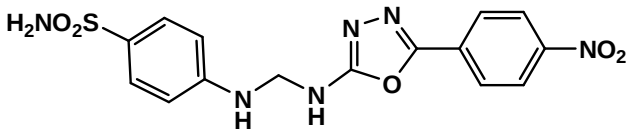
VIb.....Smylin.




```
Current Data Parameters
NAME                  Feb01-2012
EXPNO                         22
PROCNO                         1

F2 - Acquisition Parameters
Date_                   20120201
Time                       16.09
INSTRUM                    spect
PROBHD     5 mm PABBO BB-
PULPROG                     zg30
TD                         32768
SOLVENT                     DMSO
NS                             32
DS                              2
SWH                    10330.578 Hz
FIDRES                 0.315264 Hz
AQ                     1.5860212 sec
RG                           203
DW                        48.400 usec
DE                         6.50 usec
TE                        295.5 K
D1                   1.00000000 sec
TD0                            1

======== CHANNEL f1 ========
NUC1                          1H
P1                         10.65 usec
PL1                         0.00 dB
PL1W                23.53637505 W
SFO1                500.1330885 MHz

F2 - Processing parameters
SI                         32768
SF                  500.1300000 MHz
WDW                           EM
SSB                            0
LB                          0.30 Hz
GB                             0
PC                          1.00
```

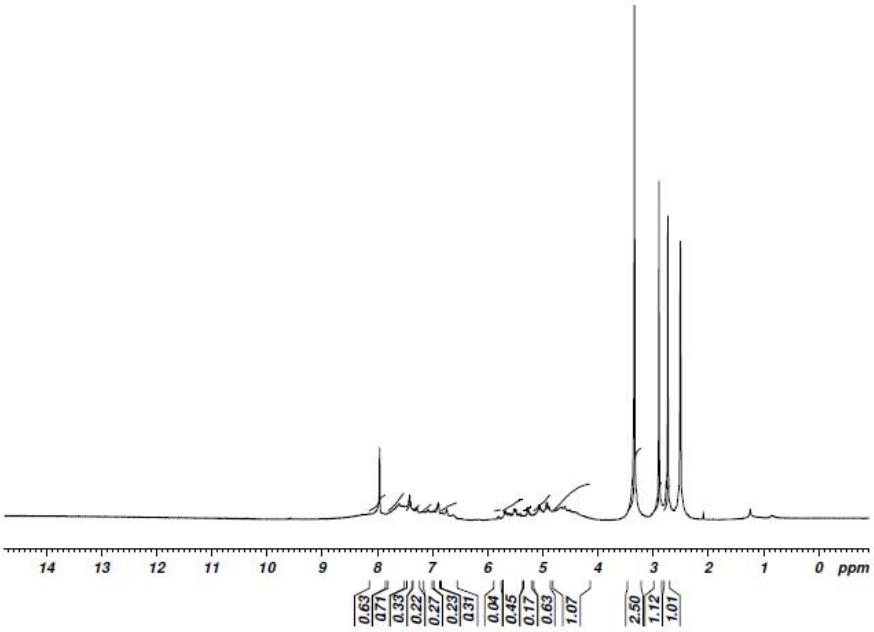
VIb.....Smylin.


5.130
5.118
5.107
3.347
2.508
5.3 5.2 5.1 5.0 4.9 4.8 4.7 4.6 4.5 4.4 4.3 4.2 4.1 4.0 3.9 3.8 3.7 3.6 3.5 3.4 3.3 3.2 3.1 3.0 2.9 2.8 2.7 2.6 2.5 ppm
2.04
1.00
1.31



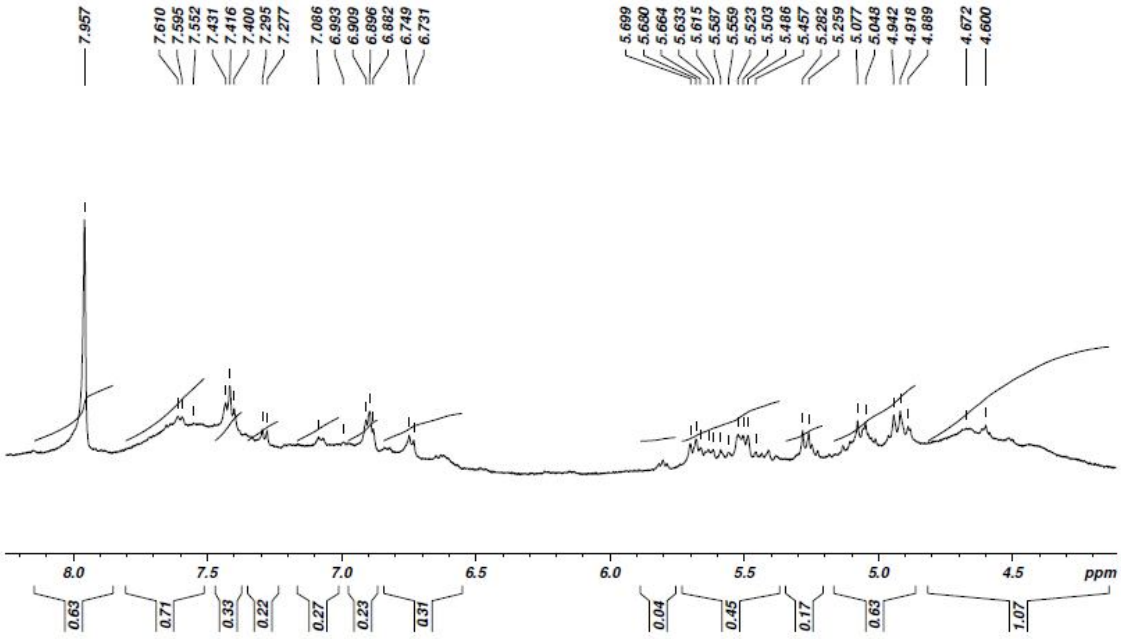
4-[({[5-(4-nitrophenyl)-1,3,4-oxadiazol-2-yl]amino}methyl)amino]benzenesulfonamide

Vle.....Smylin.

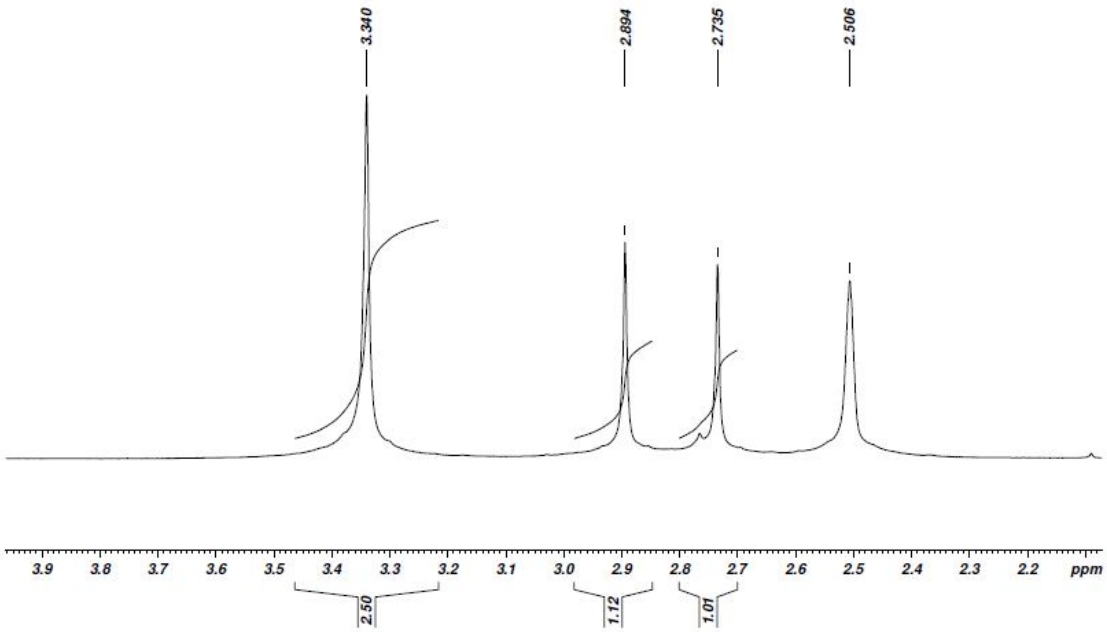


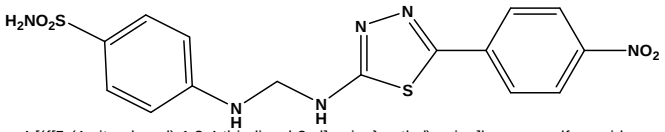

```
Current Data Parameters
NAME          Feb01-2012
EXPNO                 20
PROCNO                 1

F2 - Acquisition Parameters
Date_           20120201
Time                16.00
INSTRUM             spect
PROBHD   5 mm PABBO BB-
PULPROG              zg30
TD                  32768
SOLVENT              DMSO
NS                     32
DS                      2
SWH           10330.578 Hz
FIDRES         0.315264 Hz
AQ             1.5860212 sec
RG                    203
DW                48.400 usec
DE                 6.50 usec
TE                295.5 K
D1            1.00000000 sec
TD0                    1

====== CHANNEL f1 ======
NUC1                  1H
P1                 10.65 usec
PL1                0.00 dB
PL1W        23.53637505 W
SFO1        500.1330885 MHz

F2 - Processing parameters
SI                 32768
SF          500.1300000 MHz
WDW                   EM
SSB                    0
LB                  0.30 Hz
GB                     0
PC                  1.00
```

Vle.....Smylin.


7.957
7.610
7.595
7.552
7.431
7.416
7.400
7.295
7.277
7.086
6.993
6.909
6.896
6.882
6.749
6.731
5.699
5.680
5.664
5.633
5.615
5.587
5.559
5.523
5.503
5.486
5.457
5.282
5.259
5.077
5.048
4.942
4.918
4.889
4.672
4.600
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
ppm
0.63
0.71
0.33
0.22
0.27
0.23
0.31
0.04
0.45
0.17
0.63
1.07

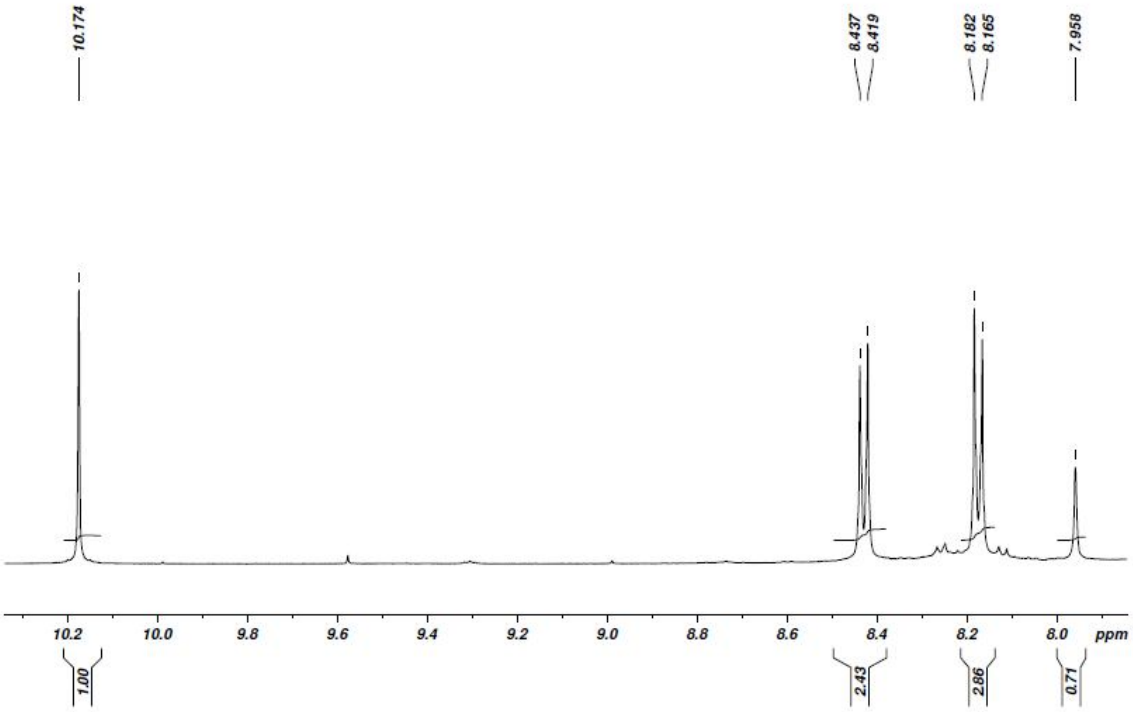
Vle.....Smylin.


3.340
2.894
2.735
2.506
3.9
3.8
3.7
3.6
3.5
3.4
3.3
3.2
3.1
3.0
2.9
2.8
2.7
2.6
2.5
2.4
2.3
2.2
ppm
2.50
1.12
1.01

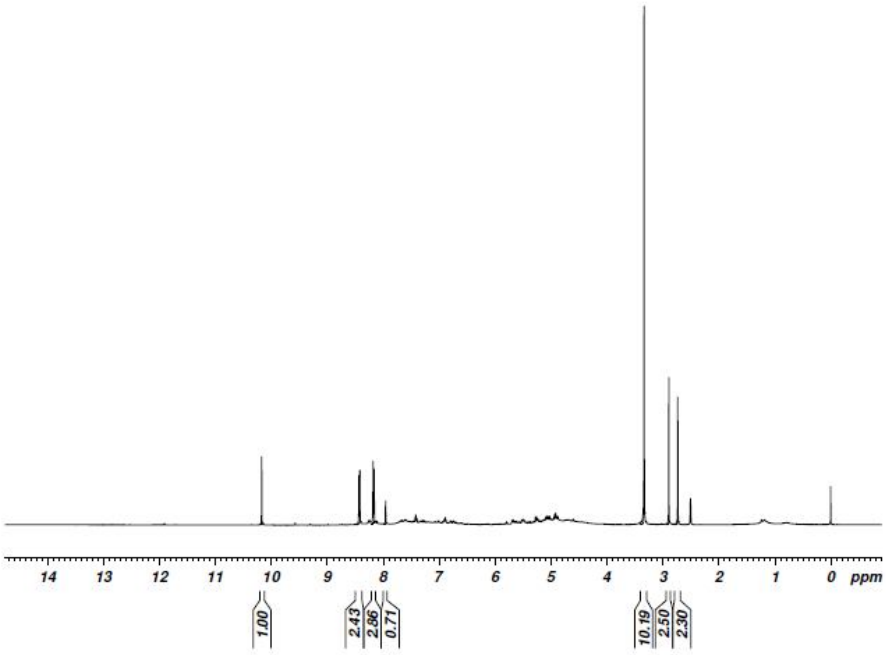


4-[({[5-(4-nitrophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulfonamide

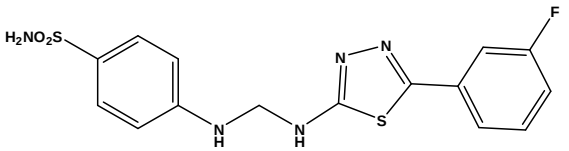
VIIId.....Smylin.



VIIId.....Smylin.

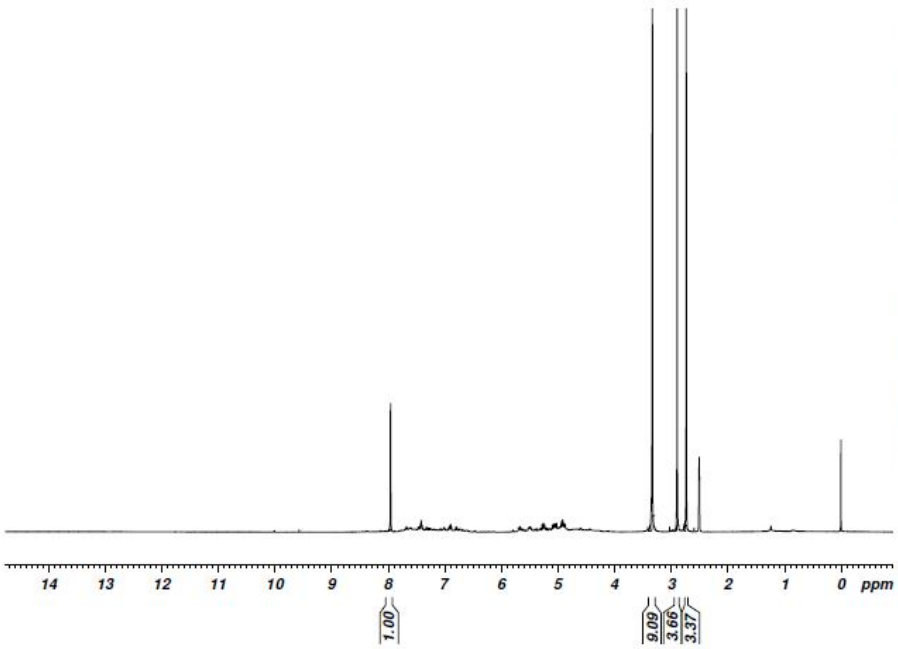



```
Current Data Parameters
NAME                  Feb16-2012
EXPNO                         17
PROCNO                         1

F2 - Acquisition Parameters
Date_                   20120216
Time                       16.26
INSTRUM                    spect
PROBHD    5 mm PABBO BB-
PULPROG                    zg30
TD                        32768
SOLVENT                    DMSO
NS                           32
DS                            2
SWH                   10330.578 Hz
FIDRES                0.315264 Hz
AQ                    1.5860212 sec
RG                          203
DW                       48.400 usec
DE                        6.50 usec
TE                       297.1 K
D1                    1.00000000 sec
TD0                            1

====== CHANNEL f1 ======
NUC1                          1H
P1                       10.65 usec
PL1                       0.00 dB
PL1W                 23.53637505 W
SFO1                 500.1330885 MHz

F2 - Processing parameters
SI                        32768
SF                   500.1300000 MHz
WDW                          EM
SSB                           0
LB                        0.30 Hz
GB                            0
PC                         1.00
```



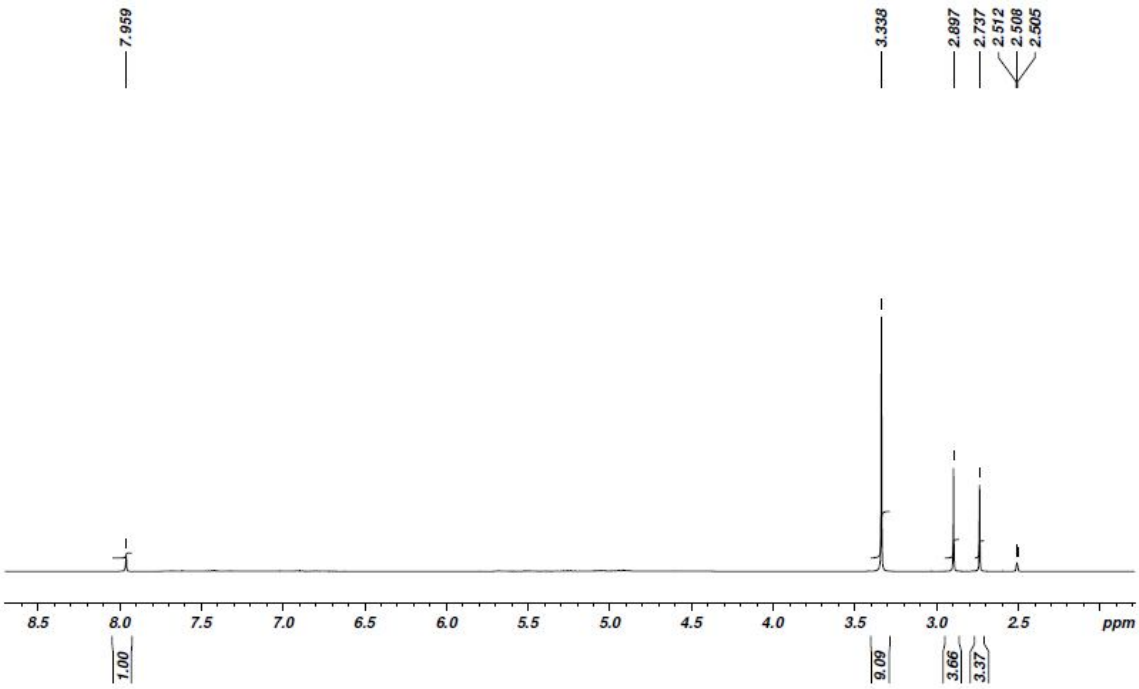
4-[({[5-(3-fluorophenyl)-1,3,4-thiadiazol-2-yl]amino}methyl)amino]benzenesulfonamide

VIIIC.....Smylin.




```
Current Data Parameters
NAME          Feb16-2012
EXPNO                 16
PROCNO                 1

F2 - Acquisition Parameters
Date_           20120216
Time               16.21
INSTRUM             spect
PROBHD   5 mm PABBO BB-
PULPROG             zg30
TD                 32768
SOLVENT             DMSO
NS                    32
DS                     2
SWH            10330.578 Hz
FIDRES         0.315264 Hz
AQ             1.5860212 sec
RG                   203
DW                48.400 usec
DE                 6.50 usec
TE                297.0 K
D1             1.00000000 sec
TD0                    1

======= CHANNEL f1 =======
NUC1                  1H
P1                10.65 usec
PL1                0.00 dB
PL1W        23.53637505 W
SFO1        500.1330885 MHz

F2 - Processing parameters
SI                 32768
SF          500.1300000 MHz
WDW                   EM
SSB                    0
LB                 0.30 Hz
GB                     0
PC                 1.00
```

VIIIC.....Smylin.



BIOLOGICAL ACTIVITY

BIOLOGICAL ACTIVITY

The newly synthesized compound were screened for their in Vitro antimicrobial activity against bacterias such *as Escherichia coli, Pseudomonas aureginosa, Rhodosporum rubrum, Vibrio cholera, Monococcus leuteus, Staphylococcus aureus, Bacillus substiles,* and *Corynebacterium typhi* etc. whereas Antifungal activity was screened by fungus such as *Candida albicans, Monococcus purpurea, Aspergillus Niger,* and *Trycophytan rubrum* etc was used.

1. Zone of inhibition (disc diffusion method)
2. Minimum inhibitory concentration (serial dilution method)

Antibacterial activity

Disc diffusion method:

Disc diffusion method was used for determination of preliminary antibacterial or antifungal activity. The disc diffusion method, the drug potency is based on measurement of the diameter of the zone of inhibition surrounding cylinder discs.

In disk diffusion method each disk was dipped into 100 micro gram concentration of synthesized compound for around 1hr. The procedure for carrying out disc diffusion method is as follows.

Preparation of inoculums:

Preparation of inoculums of bacteria was carried out by preparing Mueller Hinton Broth and transferred to test tube, keep it for sterilization in autoclave at 121□c for 15 min. then add culture of each bacteria to each tube (this step is carried out in aseptic room near laminar air flow) then keep it for incubation in incubator for 18-24 hrs at 37□c.

Procedure:

Mueller Hinton agar was prepared by adding 21gm of Mueller Hinton agar in 1000 ml of distilled water and add agar-agar 1-2 gm for solubilization.cover and keep it for sterilization in autoclave for 121□c for 15 mins. After cooling take it to aseptic room near Laminar air flow.Take a sterile Petridish and divide it into 4 or 6 quadrant. Pass the molten nutrient agar (Mueller Hinton agar) into Petridish and allow it to solidify for 10 min. Swab the organism over the molten nutrient agar. Place the disc of known concentration in a corner of each quadrant and disc of standard drug in the start. Incubate the Petridish at 37ºc for 1hrs in inverted position to allow disc diffusion as well as to half the growth of microorganism. Measure the zone of inhibition of all drugs in millimeter.

Measurement of minimum inhibitory concentration

Dilution method:

Preparation of test drug:

Serial dilution of the test Antimicrobial agent was made in 1ml of Mueller Hinton broth test series of 1-8 dilution.

Preparation of inoculums

Overnight culture grown at 37°c in Mueller Hinton broth and keep it at 37°c for 24 hrs in incubator.

Procedure:

Prepare Mueller Hinton both by formula:

Mueller Hinton	2.1g
Water	100ml

Take 8 MIC tubes ,labeled it as number 1-8 and transfer 1ml of broth to each tube, add 1ml of synthesized compound to first tube and then transfer 1ml from first tube to second ,so

similarly transfer 1ml from each tube to second tube up to 7^{th} tube and 8^{th} tube kept as controlled i.e. without drug. To all tube add 0.1ml of culture (bacterial) up to 8^{th} tubes. All the tubes were incubated at $37^{\circ}C$ for 24 hrs. After incubation observed the turbidity. The tube which shows turbidity and the tube which is before that tube and whose conc. are called as minimum inhibitory concentration.

Table no.8

code	Conc. (in µg)	Zone of inhibition(in millimeter)			
		Escherichia coli **(Gram - ve)**	***Pseudomonas. aeruginosa*** **(Gram - ve)**	***Rhodosporm rubrum*** **(Gram - ve)**	***Vibrio cholerae*** **(Gram - ve)**
3b	100	8	15	14	17
3c	100	7	13	11	8
3d	100	15	18	10	18
4b	100	11	15	7	9
4e	100	7	14	8	11
6b	100	6	16	5	10
$8a_1$	100	13	15	7	17
$8a_2$	100	12	20	23	19
$8b_1$	100	11	11	15	14
$8c_1$	100	15	13	17	16
$8d_1$	100	16	15	20	19
Sparfloxacin	100	25	20	26	25

Table no.9

code	Conc. (in µg)	Zone of inhibition(in millimeter)			
		Monococcus luteus **(Gram + ve)**	***Staphylococcus aureeus*** **(Gram + ve)**	***Bacillus substiles*** **(Gram + ve)**	***Corynebacterm typhi*** **(Gram + ve)**
3b	100	15	15	14	20
3c	100	10	9	11	11
3d	100	8	11	9	25
4b	100	14	16	7	20
4e	100	12	13	11	10
6b	100	10	8	9	9
$8a_1$	100	12	11	11	10
$8a_2$	100	18	18	14	19
$8b_1$	100	11	11	12	11
$8c_1$	100	10	10	18	12
$8d_1$	100	26	23	27	25
Sparfloxacin	100	30	25	30	30

Table no.10

Escherischia coli								
Test tube no.	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**
concentration	**100**	**50**	**25**	**12.5**	**6.25**	**3.125**	**1.5625**	**Controlled**
3b	-	-	+	+	+	+	+	+
3c	-	-	+	+	+	+	+	+
3d	-	+	+	+	+	+	+	+
4b	-	+	+	+	+	+	+	+
4e	-	-	-	+	+	+	+	+
6b	-	-	+	+	+	+	+	+
$8a_1$	-	-	-	-	-	+	+	+
$8a_2$	-	-	-	-	+	+	+	+
$8b_1$	-	-	-	-	+	+	+	+
$8c_1$	-	-	-	-	-	+	+	+
$8d_1$	-	-	-	-	-	+	+	+

Table no.11

Pseudomonas aeruginosa								
Test tube no.	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**
concentration	**100**	**50**	**25**	**12.5**	**6.25**	**3.125**	**1.5625**	**Controlled**
3b	-	-	-	-	-	+	+	+
3c	-	-	-	-	+	+	+	+
3d	-	-	-	-	-	+	+	+
4b	-	-	-	-	+	+	+	+
4e	-	-	-	+	+	+	+	+
6b	-	-	-	-	+	+	+	+
$8a_1$	-	-	-	-	-	+	+	+
$8a_2$	-	-	-	-	-	+	+	+
$8b_1$	-	-	-	-	+	+	+	+
$8c_1$	-	-	+	+	+	+	+	+
$8d_1$	-	-	-	-	-	+	+	+

Table no.12

Rhodosporum ruprum								
Test tube no.	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**
concentration	**100**	**50**	**25**	**12.5**	**6.25**	**3.125**	**1.5625**	**Controlled**
3b	-	-	-	-	+	+	+	+
3c	-	-	-	+	+	+	+	+
3d	-	-	+	+	+	+	+	+
4b	-	+	+	+	+	+	+	+
4e	-	-	-	+	+	+	+	+
6b	-	-	+	+	+	+	+	+
$8a_1$	-	-	+	+	+	+	+	+
$8a_2$	-	-	-	-	+	+	+	+
$8b_1$	-	-	-	-	-	+	+	+
$8c_1$	-	-	-	-	-	+	+	+
$8d_1$	-	-	-	-	-	+	+	+

Table no.13

Vibryo cholerae								
Test tube no.	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**
concentration	**100**	**50**	**25**	**12.5**	**6.25**	**3.125**	**1.5625**	**Controlled**
3b	-	-	-	-	-	-	+	+
3c	-	-	+	+	+	+	+	+
3d	-	-	-	-	-	-	+	+
4b	-	+	+	+	+	+	+	+
4e	-	-	-	+	+	+	+	+
6b	-	-	+	+	+	+	+	+
$8a_1$	-	-	-	-	-	+	+	+
$8a_2$	-	-	-	-	-	-	+	+
$8b_1$	-	-	-	-	+	+	+	+
$8c_1$	-	-	-	-	-	+	+	+
$8d_1$	-	-	-	-	-	-	+	+

Table no.14

Monococcus leuteus								
Test tube no.	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**
concentration	**100**	**50**	**25**	**12.5**	**6.25**	**3.125**	**1.5625**	**Controlled**
3b	-	-	-	-	-	+	+	+
3c	-	-	+	+	+	+	+	+
3d	-	-	-	-	-	+	+	+
4b	-	-	-	-	-	+	+	+
4e	-	-	+	+	+	+	+	+
6b	-	+	+	+	+	+	+	+
$8a_1$	-	-	+	+	+	+	+	+
$8a_2$	-	-	-	-	-	-	+	+
$8b_1$	-	-	-	+	+	+	+	+
$8c_1$	-	-	+	+	+	+	+	+
$8d_1$	-	-	-	-	-	-	+	+

Table no.15

Staphylococcus aureus								
Test tube no.	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**
concentration	**100**	**50**	**25**	**12.5**	**6.25**	**3.125**	**1.5625**	**Controlled**
3b	-	-	-	-	-	-	+	+
3c	-	-	-	+	+	+	+	+
3d	-	-	-	+	+	+	+	+
4b	-	-	-	-	-	-	+	+
4e	-	-	-	-	+	+	+	+
6b	-	-	+	+	+	+	+	+
$8a_1$	-	-	-	-	+	+	+	+
$8a_2$	-	-	-	-	-	+	+	+
$8b_1$	-	-	-	-	+	+	+	+
$8c_1$	-	-	-	-	+	+	+	+
$8d_1$	-	-	-	-	-	-	+	+

Table no.16

Bacillus substiles								
Test tube no.	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**
concentration	**100**	**50**	**25**	**12.5**	**6.25**	**3.125**	**1.5625**	**Controlled**
3b	-	-	-	-	-	-	+	+
3c	-	-	-	-	-	+	+	+
3d	-	-	-	+	+	+	+	+
4b	-	+	+	+	+	+	+	+
4e	-	-	-	-	-	+	+	+
6b	-	-	+	+	+	+	+	+
$8a_1$	-	-	-	+	+	+	+	+
$8a_2$	-	-	-	-	-	+	+	+
$8b_1$	-	-	-	-	-	+	+	+
$8c_1$	-	-	-	-	-	-	+	+
$8d_1$	-	-	-	-	-	-	+	+

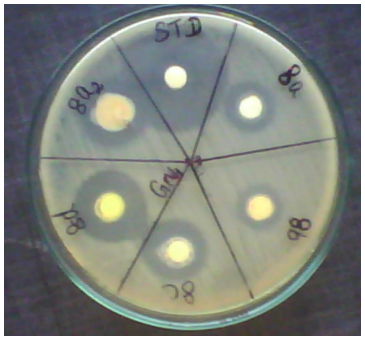
Table no.17

Corneybacterium typhi								
Test tube no.	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**
concentration	**100**	**50**	**25**	**12.5**	**6.25**	**3.125**	**1.5625**	**Controlled**
3b	-	-	-	-	-	+	+	+
3c	-	-	-	-	+	+	+	+
3d	-	-	-	-	-	-	+	+
4b	-	-	-	-	-	+	+	+
4e	-	-	-	-	+	+	+	+
6b	-	-	+	+	+	+	+	+
$8a_1$	-	-	-	+	+	+	+	+
$8a_2$	-	-	-	-	+	+	+	+
$8b_1$	-	-	-	-	+	+	+	+
$8c_1$	-	-	-	-	-	+	+	+
$8d_1$	-	-	-	-	-	-	+	+

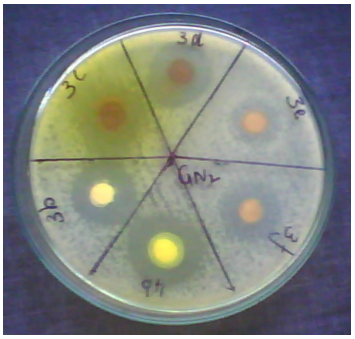
(-) Indicates = No turbidity,

(+) indicates = Turbidity,

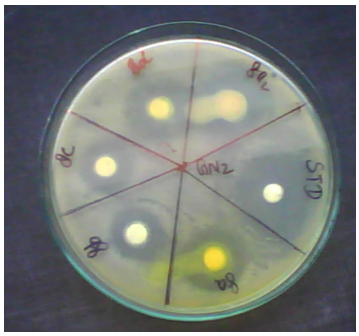
Solvent = Dimethylsulfoxide



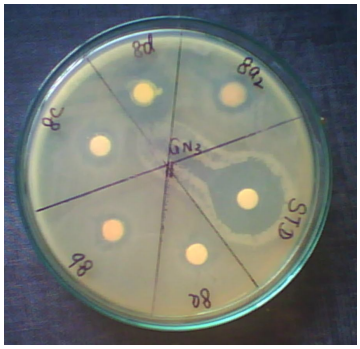
Antibacterial evaluation of compounds against ***E.coli***



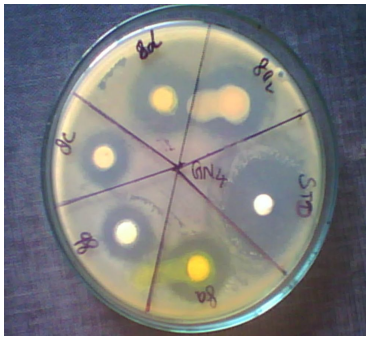
Antibacterial evaluation of compounds against ***P. aeruginosa***



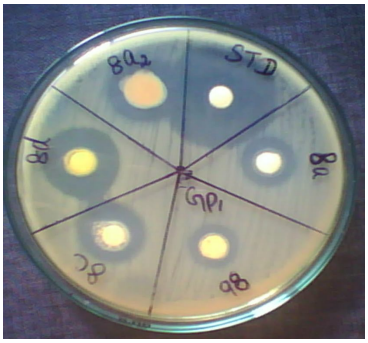
Antibacterial evaluation of compounds against ***P. aeruginosa***



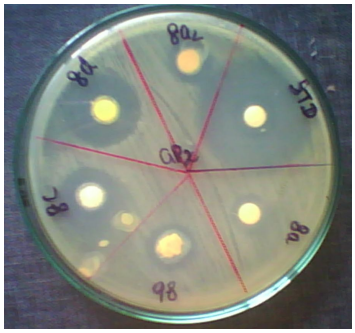
Antibacterial evaluation of compounds against ***R.rubrum***



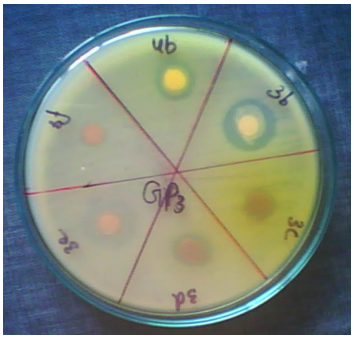
Antibacterial evaluation of compounds against ***V.Cholerae***



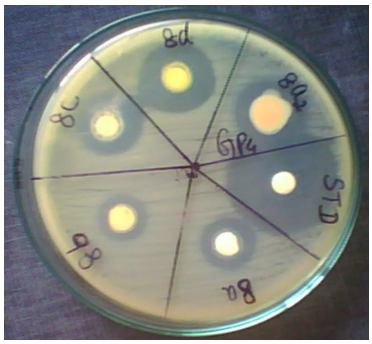
Antibacterial evaluation of compounds against ***M.luteus***



Antibacterial evaluation of compounds against ***S.aureus***



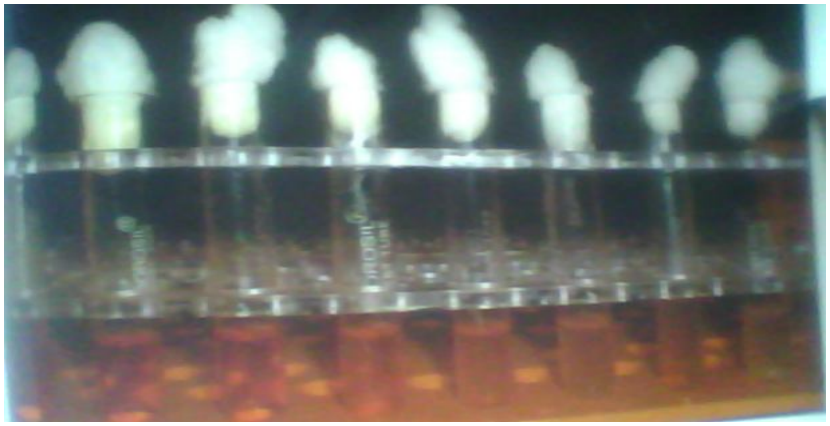
Antibacterial evaluation of compounds against ***B.substilus***



Antibacterial evaluation of compounds against ***C.typhi***

Minimum inhibitory concentration against *E.coli*

Minimum inhibitory concentration against *P. aeruginosa*



Minimum inhibitory concentration against *R.rubrum*

Minimum inhibitory concentration against *V.Cholerae*

Minimum inhibitory concentration against *M.luteus*

Minimum inhibitory concentration against *S.aureus*

Minimum inhibitory concentration against *B.substiles*

Minimum inhibitory concentration against *C.typhi*

Antifungal activity:

Disc diffusion method

Disc diffusion method was used for determination of preliminary antibacterial or antifungal activity. In disk diffusion method each disk was dipped into 100 micro gram concentration of synthesized compound for around 1hr. the procedure for carrying out disc diffusion method is as follows:

Preparation of inoculums:

Preparation of inoculums of fungus was carried out by preparing subarrod dextrose Broth and transferred to test tube, keep it for sterilization in autoclave at 121°c for 15 min. then add culture of each fungi to each tube (this step is carried out in aseptic room near laminar air flow) then keep it for incubation in incubator for 24-48 hrs. at 29°c.

Procedure:

Subarrod Dextrose Broth was prepared by adding 1g of peptone and 4g of dextrose in 100 ml of distilled water. Cover and keep it for sterilization in autoclave for 121°c for 15 min. After cooling take it to aseptic room near Laminar air flow .Take a sterile Petridish and divide it into. 4 or 6 quadrant. Pass the Subarrod Dextrose Broth into Petridish and allow it to solidify for 10 min. Swab the organism over the molten nutrient agar. Place the disc of known concentration in a corner of each quadrant and disc of standard drug in the start. Incubate the Petridish at 37°c for 1hrs in inverted position to allow disc diffusion as well as to half the growth of microorganism. Incubate the Petridish at 29°c for 24-48 hrs. in inverted position. Measure the zone of inhibition of all drugs in millimeter.

Measurement of minimum inhibitory concentration

Dilution method:

Preparation of test drug:

Serial dilution of the test Antimicrobial agent was made in 1ml of Subarrod Dextrose Broth test series of 1-8 dilution.

Preparation of inoculums

Overnight culture grown at 29°C in Subarrod Dextrose Broth and keep it at 29°C for 24-48 hrs. in incubator.

Procedure:

Prepare Subarrod Dextrose Broth by formula:

Peptone	1g
Dextrose	4g
Water	100ml

Take 8 MIC tubes, labeled it as number 1-8 and transfer 1ml of broth to each tube. add .then add 1ml of drug to first tube then transfer 1ml from first tube to second so similarly transfer 1ml from each tube to second tube up to 7^{th} tube and 8^{th} tube kept as controlled i.e. without drug. To all tube add 0.1ml of culture (fungi) up to 8^{th} tubes. All the tubes were incubated at $29^{\circ}C$ for 24-48 hrs. After incubation observed the turbidity. The tube which shows turbidity the tube which is before that tube and whose conc. are called as minimum inhibitory concentration.

Table no.18

Compound Code	Concentration (in µg)	Zone of inhibition in millimeter			
		Candida albicans	***Monococcus purpurea***	***Aspergillus niger***	***Trycophytan rubrum***
3b	100 µg	6	8	8	5
3c	100 µg	8	8	6	6
3d	100 µg	7	7	5	8
4b	100 µg	6	5	9	8
4e	100 µg	11	9	7	15
6b	100 µg	7	6	16	10
$8a_1$	100 µg	10	9	13	13
$8a_2$	100 µg	17	11	13	18
$8b_1$	100 µg	14	11	11	14
$8c_1$	100 µg	11	10	10	14
$8d_1$	100 µg	13	10	11	12
Clotrimazole	100 µg	15	12	19	20

Table no.19

Candida albicans								
Test tube no.		**2**	**3**	**4**	**5**	**6**	**7**	**8**
concentration	**100**	**50**	**25**	**12.5**	**6.25**	**3.125**	**1.5625**	**Controlled**
3b	-	-	-	+	+	+	+	+
3c	-	-	+	+	+	+	+	+
3d	-	-	+	+	+	+	+	+
4b	-	+	+	+	+	+	+	+
4e	-	-	-	-	-	+	+	+
6b	-	-	+	+	+	+	+	+
$8a_1$	-	-	-	+	+	+	+	+
$8a_2$	-	-	-	-	+	+	+	+
$8b_1$	-	-	-	-	+	+	+	+
$8c_1$	-	-	-	-	+	+	+	+
$8d_1$	-	-	-	-	-	+	+	+

Table no.20

Monococcus purpurea								
Test tube no.	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**
concentration	**100**	**50**	**25**	**12.5**	**6.25**	**3.125**	**1.5625**	**Controlled**
3b	-	-	-	-	+	+	+	+
3c	-	-	-	-	+	+	+	+
3d	-	-	-	+	+	+	+	+
4b	-	+	+	+	+	+	+	+
4e	-	-	-	-	-	+	+	+
6b	-	-	+	+	+	+	+	+
$8a_1$	-	-	-	+	+	+	+	+
$8a_2$	-	-	-	-	-	+	+	+
$8b_1$	-	-	-	-	+	+	+	+
$8c_1$	-	-	-	-	+	+	+	+
$8d_1$	-	-	-	-	-	+	+	+

Table no.21

Aspergillus niger								
Test tube no.	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**
concentration	**100**	**50**	**25**	**12.5**	**6.25**	**3.125**	**1.5625**	**Controlled**
3b	-	-	-	-	+	+	+	+
3c	-	-	-	-	-	+	+	+
3d	-	-	-	-	-	+	+	+
4b	-	-	-	-	-	+	+	+
4e	-	-	-	-	-	-	+	+
6b	-	-	-	-	-	-	+	+
$8a_1$	-	-	-	-	-	+	+	+
$8a_2$	-	-	-	-	-	+	+	+
$8b_1$	-	-	-	-	-	-	+	+
$8c_1$	-	-	-	-	-	-	+	+
$8d_1$	-	-	-	-	-	+	+	+

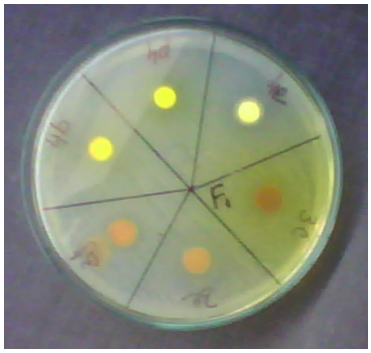
Table no.22

Monococcus rubrum								
Test tube no.	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**
concentration	**100**	**50**	**25**	**12.5**	**6.25**	**3.125**	**1.5625**	**Controlled**
3b	-	-	-	-	+	+	+	+
3c	-	-	-	-	-	+	+	+
3d	-	-	-	-	-	+	+	+
4b	-	-	-	-	-	+	+	+
4e	-	-	-	-	-	-	+	+
6b	-	-	-	-	-	-	+	+
$8a_1$	-	-	-	-	-	+	+	+
$8a_2$	-	-	-	-	-	+	+	+
$8b_1$	-	-	-	-	-	+	+	+
$8c_1$	-	-	-	-	-	-	+	+
$8d_1$	-	-	-	-	-	+	+	+

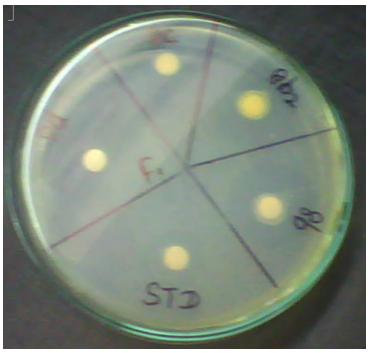
(-) Indicates = No turbidity,

(+) indicates = Turbidity,

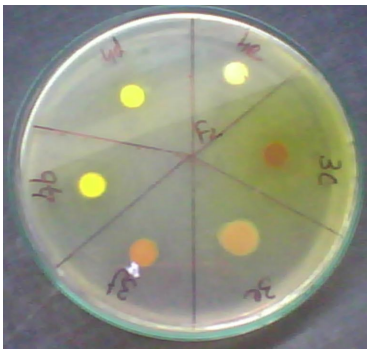
Solvent = Dimethylsulfoxide



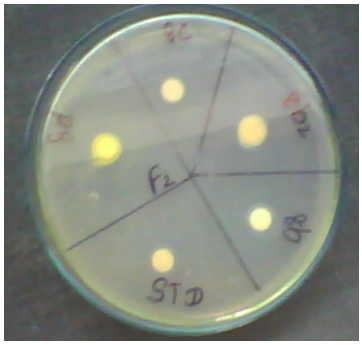
Antifungal evaluation of compounds against *C.albicans*



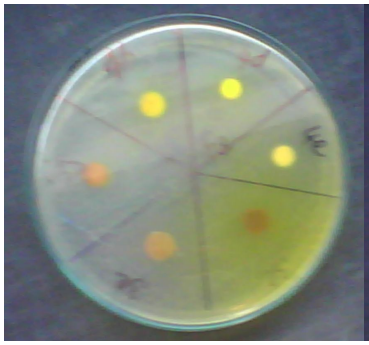
Antifungal evaluation of compounds against *C.albicans*



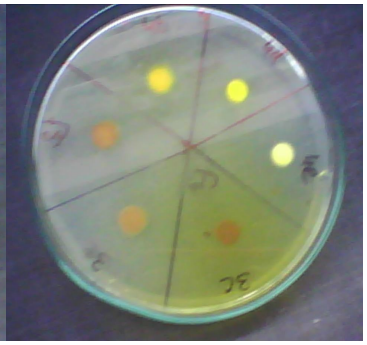
Antifungal evaluation of compounds against *M.purpurea*



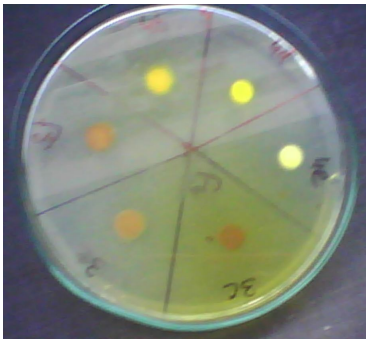
Antifungal evaluation of compounds against *M.purpurea*



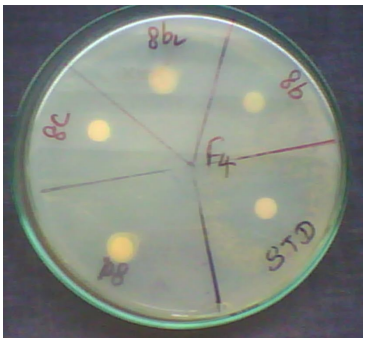
Antifungal evaluation of compounds against *A.niger*



Antifungal evaluation of compounds against *A.niger*



Antifungal evaluation of compounds against *M.rubrum*



Antifungal evaluation of compounds against *M.rubrum*

Minimum inhibitory concentration against *C.albicans*

Minimum inhibitory concentration against *M.purpurea*

Minimum inhibitory concentration against *A.niger*

Minimum inhibitory concentration against *M.rubrum*

IN VITRO ANTI CANCER ACTIVITY

The human cervical cancer cell line (HeLa) and mouse embryonic fibroblasts cell line (NIH 3T3) were obtained from National Centre for Cell Science (NCCS), Pune. The HeLa cells were grown in Eagles Minimum Essential Medium containing 10% fetal bovine serum (FBS) and NIH 3T3 cells were grown in Dulbeccos Modified Eagles Medium (DMEM) containing 10% FBS. All cells were maintained at 37^0C, 5% CO2, 95% air and 100% relative humidity. Maintenance cultures were passaged weekly, and the culture medium was changed twice a week.

Cell treatment procedure

The monolayer cells were detached with trypsin-ethylenediaminetetraacetic acid (EDTA) to make single cell suspensions and viable cells were counted using a hemocytometer and diluted with medium with 5% FBS to give final density of $1x10^5$ cells/ml. one hundred microlitres per well of cell suspension were seeded into 96-well plates at plating density of 10,000 cells/well and incubated to allow for cell attachment at 37^0C, 5% CO_2, 95% air and 100% relative humidity. After 24 h the cells were treated with serial concentrations of the extracts and fractions. They were initially dissolved in neat dimethylsulfoxide (DMSO) and further diluted in serum free medium to produce five concentrations. One hundred microlitres per well of each concentration was added to plates to obtain final concentrations of 100, 10, 1.0 and 0.1 μM. The final volume in each well was 200 μl and the plates were incubated at 37^0C, 5% CO_2, 95% air and 100% relative humidity for 48h. The medium containing without samples were served as control. Triplicate was maintained for all concentrations.

MTT assay

MTT is a yellow water soluble tetrazolium salt. A mitochondrial enzyme in living cells, succinate-dehydrogenase, cleaves the tetrazolium ring, converting the MTT to an

insoluble purple formazan. Therefore, the amount of formazan produced is directly proportional to the number of viable cells.After 48h of incubation, 15µl of MTT (5mg/ml) in phosphate buffered saline (PBS) was added to each well and incubated at 37^0C for 4h. The medium with MTT was then flicked off and the formed formazan crystals were solubilized in 100µl of DMSO and then measured the absorbance at 570 nm using micro plate reader. The % cell inhibition was determined using the following formula.

% cell Inhibition = 100- Abs (sample)/Abs (control) x100

Nonlinear regression graph was plotted between % Cell inhibition and Log_{10} concentration and IC_{50} was determined using GraphPad Prism software.[67,68]

Table no.23 Absorbance of various concentration of samples at 570 nm in HELA cell line.

Code	Conc (µM)	0.1	1	10	100	Cont	IC_{50} (µM)
1b	ABS	0.414	0.386	0.368	0.276	0.426	
		0.413	0.384	0.373	0.288	0.391	
		0.395	0.404	0.372	0.277	0.415	
	Avg	**0.407333**	**0.391333**	**0.371**	**0.280333**	**0.41066**	
	% cell inhibition	0.811688	4.707792	9.659091	31.73701		**> 100**
4e	ABS	0.39	0.388	0.37	0.337	0.426	
		0.404	0.4	0.383	0.326	0.391	
		0.41	0.398	0.366	0.318	0.415	
	Avg	**0.401333**	**0.395333**	**0.373**	**0.327**	**0.410667**	
	% cell inhibition	2.272727	3.733766	9.172078	20.37338		**> 100**

Table no.24 Absorbance of various concentration of samples at 570 nm in normal cell line.

Code	Conc (µM)	0.1	1	10	100	Cont	IC_{50} (µM)
1b	ABS	0.145	0.239	0.219	0.155	0.245	
		0.241	0.241	0.215	0.164	0.248	
		0.244	0.238	0.211	0.17	0.247	
	Avg	**0.21**	**0.239333**	**0.215**	**0.163**	**0.246667**	
	% cell inhibition	14.86486	2.972973	12.83784	33.91892		**> 100**
4e	ABS	0.25	0.235	0.214	0.093	0.245	
		0.243	0.242	0.211	0.116	0.248	
		0.242	0.237	0.216	0.111	0.247	
	Avg	0.245	0.238	0.213667	0.106667	0.246667	
	% cell inhibition	0.675676	3.513514	13.37838	56.75676		**> 100**

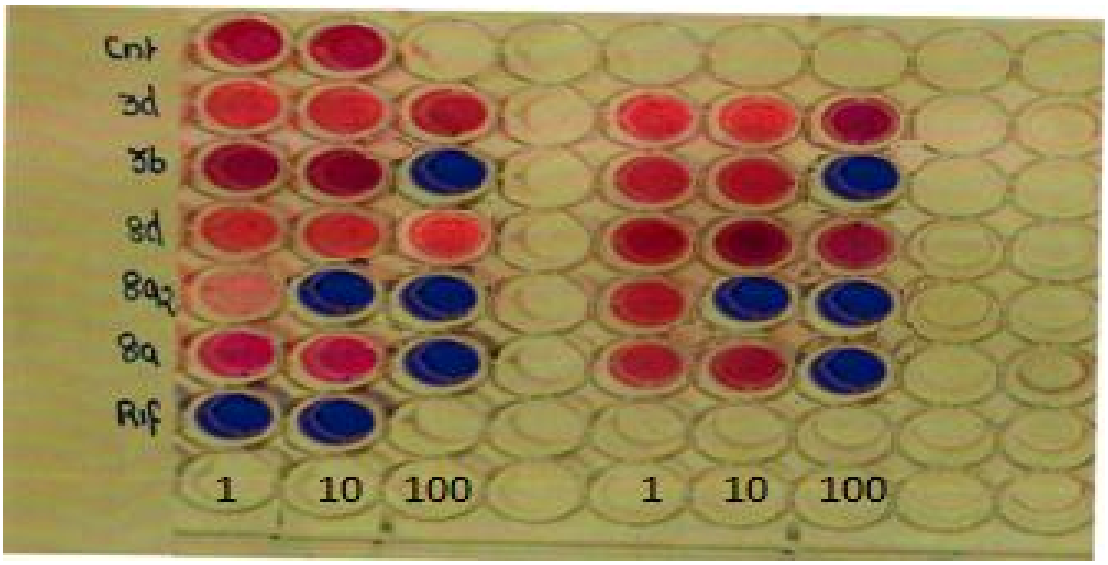
Anti-Tubercular Activity of Synthesized Compounds

The REMA(Resazurin microplate assay) plate method was performed to determine the MICs of test compounds for all the Mycobacterium isolates briefly, a 100 ml volume of Middle brook 7H9 broth (Difco, USA) was dispensed in each well of a 96-well cell culture plate (Nunc, Denmark). Test compound concentrations prepared directly in the medium were 1, 10,100 µg/ml. Perimeter wells of the plate were filled with sterile water to avoid dehydration of the medium during incubation. A standard bacterial suspension equivalent in turbidity to that of a no. 1 McFarland standard was prepared and diluted 1:20 in 7H9 broth; a100 mL inoculum was used to inoculate each well of the plate. A growth control containing no test compound and a sterile control without inoculum were also included. Plates were sealed and incubated at 37ºC for 1 week. Twenty-five microlitres (25µL of 0.02% Resazurin (Sigma Chem. Co.) Solution was added to each well; plates were re-incubated for an additional 2 days. A change in colour from blue to pink indicated the growth of bacteria, and the MIC was read as the Minimum test compound concentration that prevented the color change in Resazurin solution.**[69,70,71]**

Assay-REMA
Test organism: Mycobacterium tuberculosis

Sr.no.	Compound code	Concentration		
		1µg/ml	10µg/ml	100µg/ml
1	3d	N	N	N
2	3b	N	N	P
3	$8d_1$	N	N	N
4	$8a_2$	N	P	P
5	$8a_1$	N	N	P
6	Rifampicin	P	P	
7	control	N	N	

N=No inhibition
P=Inhibition



RESULTS ANDDISCUSSION

RESULTS AND DISCUSSION

The title compounds were synthesized in 3 steps process:

In the first step

Synthesis of titled compound was started from substituted aromatic aldehyde, which upon reaction with semicarbazides and thiosemicarbazide in alcohol to form semicarbazone and thiosemicarbazone respectively.

In the second step

Semicarbazone and thiosemicarbazone was suspended in warm water; ferric chloride in warm water was added quantitatively, slowly with constant stirring. The contents were heated at 100°C for 2hrs. Solution was filtered and citric acid , sodium carbonate were added. The obtained mixture was divided in to four portions and each portion was neutralized with ammonia respectively. The precipitate obtained was recrystallized from alcohol.

In the third step

The 2-amino-5-substituted-1, 3, 4-oxadiazole and 2-amino-5-substituted-1,3,4-thiadiazole were undergoes mannich reaction with substituted aniline and formaldehyde to get mannich base 2-amino 5-substituted 1, 3, 4- oxadiazole and 2-amino 5-substituted 1, 3, 4- thiadiazole derivatives respectively.

Melting point

The melting point of the synthesized compounds were found by an open capillary tube method using campbell electronics model-C-CMP-1 melting point apparatus and or uncorrected.

Characterization of synthesized compounds

Solubility

The solubility of all compounds was tested by using following solvents.

Chloroform, ethanol, DMSO, DMF, acetone, etc.(soluble)

Water, CCl4, etc (insoluble)

Thin layer chromatography:

The purity of synthesized compounds and conformation of the reaction were analyzed by thin layer chromatography technique using silica gel-G (G=gypsum binder) as stationary phase, employing methanol : chloroform (1:1) as mobile phase, spots were visualized in iodine vapour.

The Rf value were calculated by using formula:

Rf value = distance travelled by sample/distance travelled by solvent front

Infrared spectra

The infrared spectrums of the synthesized compounds were given in detail. The formations of C-O-C in the compounds were clearly observed in all the spectra. the absence of COOH-peak is also a evidence for the formation of compounds

From the structure investigation, IR spectra showed the stretching frequency range between 1042-1234cm-^{1}, which evidenced the presence of C-O-C group and 676-908cm-1 which showed the presence of C-S-C group.

Nuclear magnetic resonance spectra

NMR spectra of all the compounds were given. It shows the characteristic peak for the formed compounds.

1HNMR spectra give a characteristic proton resonance shift for all the synthesized 1,3,4-oxadiazole and 1,3,4-thiadiazole derivatives which ensured the existence of aromatic or substituent proton.

Mass spectra

Mass spectra of synthesized were taken and it is giving almost same molecular weight as that of calculated molecular weight.

Biological activity:

The zone of inhibition and Minimum inhibitory concentration of the synthesized compounds were tabulated. All the synthesized compounds have showed an excellent activity against both Gram positive and negative bacteria and fungi.

Antibacterial screening

The antibacterial screening of some compounds showed an excellent zone of inhibition like compounds **$8a_1$,$8a_2$,$8b_1$,$8c_1$** and **$8d_1$** showed excellent activity against all bacterial strains. Compounds **3d** showed significant activity against *P.aureginosa, V.cholerae* and *C.typhi* and **4b** Was found to be comparable activity against *S.aures* and *C.typhi*.

Antifungal screening

The antifungal screening of synthesized compounds showed excellent zone of inhibition like**$8a_1$,$8a_2$,$8b_1$** and **$8c_1$** against all pathogenic fungal strains. The compounds **4e** and **6b** was found to be highly active against *M.rubrum* and *A.niger*. Compounds **3c, 3d** and **4b** showed moderate activity against all pathogenic strains.

Anticancer screening

The *in vitro* anticancer studies were performed for two compounds using MTT assay against HeLa cell line and normal cell line. The compounds **1b** and **4e** shown no cytotoxic activity and IC_{50} value is greater than 100 µM .

Antitubercular screening

In vitro antitubercular screening of six compounds which showed significant antibacterial and antifungal activity were studied by REMA which revealed the compounds **$8a_2$** showed significant antimycobacterial activity at a concentration of 10 µg/ml. Compounds **$8a_1$** and **3b** possessed antimycobacterial activity at a concentration of 100 µg/ml. The compound **$8d_1$** showed lack of antimycobacterial activity against *Mycobacterium tuberculosis* H37Rv strain.

SUMMARY AND CONCLUSION

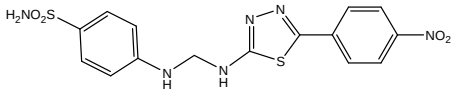
SUMMARY AND CONCLUSION

Synthesis of titled compound was started from substituted aromatic aldehyde, which upon reaction with semicarbazides and thiosemicarbazide in alcohol to form semicarbazone and thiosemicarbazone respectively. Semicarbazone and thiosemicarbazone was suspended in warm water; ferric chloride in warm water was added quantitatively, slowly constant stirring with respectively. The contents were heated at 100°C for 2hrs. Solution was filtered and citric acid sodium carbonate were added. The obtained mixture was divided in to four portions and each portion was neutralized with ammonia. The precipitate obtained was recrystallized from alcohol. The 2-amino-5-substituted-1,3,4-oxadiazole and 2-amino-5-substituted1,3,4-thiadiazole respectively were undergoes mannich reaction with substituted aniline and formaldehyde to get mannich base 2-amino 5-substituted 1,3,4-oxadiazole derivatives and 2-amino 5-substituted-1,3,4- thiadiazole derivatives respectively.

The melting point and Rf value of the synthesized compounds indicating the physical properties and purity of compounds.

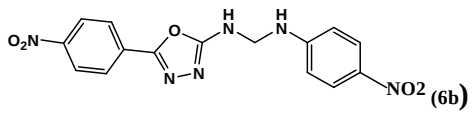
The structure of the synthesized compounds has been analyzed by Infrared spectra, NMR spectral studies and Mass spectral data's. The data's of analytical spectra were correlating with structures of synthesized compounds.

- The compound 4-[({[5-(-4-nitrophenyl)-1,3,4-thiadiazol-2yl]amino}methyl) amino]benzene sulphonamide **($8d_1$)** showed could serve a novel template for bacterial infection chemotheraphy.



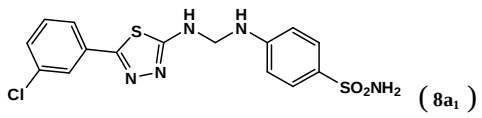
($8d_1$)

- The compound N-[5-(4-nitrophenyl)-1,3,4-oxadiazol-2-yl]-N1-nitrophenylmethane diamine **(6b)** may be potential candidate for new antifungal agent.

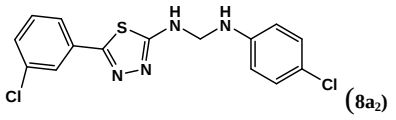


(6b)

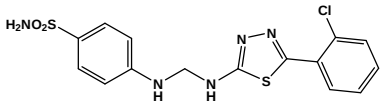
- The compounds **($8a_1$, $8a_2$, $8b_1$,** and **$8c_1$)** exhibited significant antibacterial and antifungal activities.



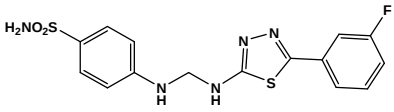
($8a_1$)



($8a_2$)

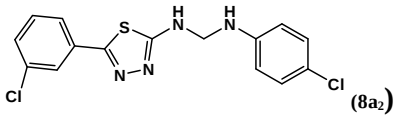


($8b_1$)



($8c_1$)

- Two compounds **(1b, 4e)** were tested against *HeLa,* normal cell and IC_{50} value is found to be 74.55 µM and the synthesized compounds show no cytotoxicity and the compound will be safe for anticancer activity.
- Compound N-(4-chlorophenyl)-N^1-[5-(3-chlorophenyl)-1,3,4-thiadiazol-2-yl)methane diamine **($8a_2$)** showed significant antimycobacterial activity at a concentration of 10 µg/ml against *Mycobacterium tuberculosis* H37Rv strain.



($8a_2$)

The entire study reveals that there is wide scope of modifications possible for 1,3,4-oxadiazole and 1,3,4-thiadiazole ring system. Oxadiazole and thiadiazole ring system could be incorporated into many more ring systems which itself have their own activity and could lead to more potent and highly active compounds. The emphasis should be laid on some other activities also such as Antialzheimer, Anticonvulsant, Antineoplastic, Antiviral, Pesticidal, Anti-inflammatory etc. Many new methods could be developed by using different solvents and substituent's. Recent interest was shown by Synthesizing the 1,3,4-oxadiazoles and 1,3,4-thiadiazoles by Microwave synthesis.

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